considered 6/20/03 MCF


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=> fil capl; d que l7
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L1          631 SEA FILE=CAPLUS ABB=ON  BRENNER S?/AU
L4        17960 SEA FILE=CAPLUS ABB=ON  LIBRAR?/OBI
L6       215485 SEA FILE=CAPLUS ABB=ON  (DNA OR NUCLEIC ACID#)/CW
L7           11 SEA FILE=CAPLUS ABB=ON  L1 AND L4 AND L6

=> fil biotechno; d que l33; d que l37

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FILE LAST UPDATED: 28 FEB 2003      <20030228/UP>
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L28          186 SEA FILE=BIOTECHNO ABB=ON  BRENNER S?/AU
L29        33609 SEA FILE=BIOTECHNO ABB=ON  LIBRAR?
L30           23 SEA FILE=BIOTECHNO ABB=ON  L28 AND L29
L31        95287 SEA FILE=BIOTECHNO ABB=ON  HETERO?
L32         7229 SEA FILE=BIOTECHNO ABB=ON  ?DUPLEX?
L33            1 SEA FILE=BIOTECHNO ABB=ON  L30 AND (L31 OR L32)


L28          186 SEA FILE=BIOTECHNO ABB=ON  BRENNER S?/AU
L29        33609 SEA FILE=BIOTECHNO ABB=ON  LIBRAR?
L30           23 SEA FILE=BIOTECHNO ABB=ON  L28 AND L29
L34       366312 SEA FILE=BIOTECHNO ABB=ON  DNA OR ?NUCLEIC ACID#
L36        32221 SEA FILE=BIOTECHNO ABB=ON  ?NUCLEASE?
L37            2 SEA FILE=BIOTECHNO ABB=ON  L30 AND L34 AND L36

=> s l33 or l37

L115           3 L33 OR L37
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=> fil wpids; d que l65

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L56          57 SEA FILE=WPIDS ABB=ON  BRENNER S?/AU
L57       13227 SEA FILE=WPIDS ABB=ON  LIBRAR?
L61       68152 SEA FILE=WPIDS ABB=ON  DNA OR ?NUCLEIC ACID#
L65           7 SEA FILE=WPIDS ABB=ON  L56 AND L57 AND L61


=> fil biotechds;d que l83

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L73          48 SEA FILE=BIOTECHDS ABB=ON  BRENNER S?/AU
L74        9646 SEA FILE=BIOTECHDS ABB=ON  LIBRAR?
L75       12955 SEA FILE=BIOTECHDS ABB=ON  HETERO?
L76        1136 SEA FILE=BIOTECHDS ABB=ON  DUPLEX?
L77         343 SEA FILE=BIOTECHDS ABB=ON  HETERODUPLEX?
L78       96580 SEA FILE=BIOTECHDS ABB=ON  DNA OR (NUCLEIC OR DEOXYRIBONUCLEIC)
                (W)ACID#
L79        5000 SEA FILE=BIOTECHDS ABB=ON  ENDONUCLEASE# OR ENDO NUCLEASE#
L80         212 SEA FILE=BIOTECHDS ABB=ON  (EXO OR EXONUCLEASE#)(1W)(III OR 3)

L81        3171 SEA FILE=BIOTECHDS ABB=ON  SINGLESTRAND? OR SINGLE STRAND?
L83           7 SEA FILE=BIOTECHDS ABB=ON  L73 AND L74 AND L78 AND ((L75 OR
                L76 OR L77) OR (L79 OR L80 OR L81))
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=> fil biosis; d que l103

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L94         566 SEA FILE=BIOSIS ABB=ON  BRENNER S?/AU
L95       45266 SEA FILE=BIOSIS ABB=ON  LIBRAR?
L96      717927 SEA FILE=BIOSIS ABB=ON  DNA OR ?NUCLEIC ACID#
L97      312200 SEA FILE=BIOSIS ABB=ON  HETERO?
L98       15293 SEA FILE=BIOSIS ABB=ON  DUPLEX?
L99        3106 SEA FILE=BIOSIS ABB=ON  HETERODUPLEX?
L100      24889 SEA FILE=BIOSIS ABB=ON  ENDONUCLEASE# OR ENDO NUCLEASE#
L101       1032 SEA FILE=BIOSIS ABB=ON  (EXO OR EXONUCLEASE#)(1W)(III OR 3)
L102      28579 SEA FILE=BIOSIS ABB=ON  SINGLESTRAND? OR SINGLE STRAND?
L103          2 SEA FILE=BIOSIS ABB=ON  L94 AND L95 AND L96 AND (L97 OR L98 OR
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PROCESSING COMPLETED FOR L65
L116         22 DUP REM L115 L83 L7 L103 L65 (8 DUPLICATES REMOVED)
                ANSWERS '1-3' FROM FILE BIOTECHNO
                ANSWERS '4-8' FROM FILE BIOTECHDS
                ANSWERS '9-19' FROM FILE CAPLUS
                ANSWER '20' FROM FILE BIOSIS
                ANSWERS '21-22' FROM FILE WPIDS

=> d ibib ab 1-22

L116  ANSWER 1 OF 22  BIOTECHNO  COPYRIGHT 2003 Elsevier Science B.V.DUPLICATE
ACCESSION NUMBER:        2000:30415296    BIOTECHNO
TITLE:                   Gene expression analysis by massively parallel
                         signature sequencing (MPSS) on microbead arrays
```

AUTHOR: **Brenner S.**; Johnson M.; Bridgham J.; Golda G.; Lloyd D.H.; Johnson D.; Luo S.; McCurdy S.; Foy M.; Ewan M.; Roth R.; George D.; Eletr S.; Albrecht G.; Vermaas E.; Williams S.R.; Moon K.; Burcham T.; Pallas M.; DuBridge R.B.; Kirchner J.; Fearon K.; Mao J.-I.; Corcoran K.
CORPORATE SOURCE: S. Brenner, Lynx Therapeutics, Inc., 25861 Industrial Blvd., Hayward, CA 94545, United States. E-mail: sbrenner@lynxgen.com
SOURCE: Nature Biotechnology, (2000), 18/6 (630-634), 17 reference(s)
CODEN: NABIFO ISSN: 1087-0156
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
SUMMARY LANGUAGE: English

AB We describe a novel sequencing approach that combines non-gel-based signature sequencing with in vitro cloning of millions on templates on separate 5 .mu.m diameter microbeads. After constructing a microbead **library** of **DNA** templates by in vitro cloning, we assembled a planar array of a million template-containing microbeads in a flow cell at a density greater than 3 x 10.sup.6 microbeads/cm.sup.2. Sequences of the free ends of the cloned templates on each microbead were then simultaneously analyzed using a fluorescence-based signature sequencing method that does not require **DNA** fragment separation. Signature sequences of 16-20 bases were obtained by repeated cycles of enzymatic cleavage with a type IIs restriction **endonuclease**, adaptor ligation, and sequence interrogation by encoded hybridization probes. The approach was validated by sequencing over 269,000 signatures from two cDNA **libraries** constructed from a fully sequenced strain of Saccharomyces cerevisiae, and by measuring gene expression levels in the human cell line THP-1. The approach provides an unprecedented depth of analysis permitting application of powerful statistical techniques for discovery of functional relationships among genes, whether known or unknown beforehand, or whether expressed at high or very low levels.

L116 ANSWER 2 OF 22 BIOTECHNO COPYRIGHT 2003 Elsevier Science B.V.DUPLICATE
ACCESSION NUMBER: 1991:21109638 BIOTECHNO
TITLE: Canonical ordered cosmid **library** of the symbiotic plasmid of Rhizobium species NGR234
AUTHOR: Perret X.; Broughton W.J.; **Brenner S.**
CORPORATE SOURCE: Laboratoire de Biologie Moleculaire des Plantes Superieures, Universite de Geneve, 1 chemin de l'Imperatrice, 1292 Chambesy, Geneva, Switzerland.
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (1991), 88/5 (1923-1927)
CODEN: PNASA6 ISSN: 0027-8424
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Many of the bacterial genes involved in nodulation (nod) and nitrogen fixation (nif) are dispersed over the 500-kilobase plasmid pNGR234a of the broad host-range Rhizobium species NGR234. As a first step toward generating a complete physical and genetic map of the plasmid, a full overlapping collection of cosmids was derived from a total genomic **library**. Clones were aligned by combining fingerprinting, hybridization, and pulsed-field gel electrophoresis data. Symbiotic loci were localized by probing a representative set of cosmids with both homologous and **heterologous** genes. nodABC, nodD1, nodD2, nodSU, nolB, and region II are widely dispersed over pNGR234a, while the two functional copies of nifKDH are separated by only 28 kilobases.

differentially expressed genes, particularly those expressed rarely. It is useful in e.g. pest control, therapeutics, drug design, etc. (107pp)

L116 ANSWER 5 OF 22 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI
ACCESSION NUMBER: 1999-03531 BIOTECHDS
TITLE: Constructing a high resolution physical map of a polynucleotide;
using two restriction **endonuclease** enzymes with massively parallel signature sequencing for mapping genomic **DNA** and disease-related genes
AUTHOR: **Brenner S**
PATENT ASSIGNEE: Lynx-Ther.
LOCATION: Hayward, CA, USA.
PATENT INFO: WO 9900519 7 Jan 1999
APPLICATION INFO: WO 1998-US13335 25 Jun 1998
PRIORITY INFO: US 1997-884189 27 Jun 1997
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 1999-095762 [08]
AB A method of mapping a polynucleotide is claimed and comprises: providing a first population of restriction fragments (RFs) by digesting a **DNA** sequence with a first restriction **endonuclease** (RE) (each RF being attached to discrete regions on a solid surface); sequencing (by massively parallel signature sequencing) the RF free ends; digesting the RFs with a second RE, and then sequencing each RF in the first population; sequencing each RF free end in the second population; digesting the RFs in the second population with the first RE to generate new free ends to form an ordered pair of nucleotide sequences for each RF in the second population; and assembling a map of the **DNA** sequence by overlapping sequences from the first and second populations. Also claimed is a method of ordering restriction fragments from a **DNA** sequence. The method is useful for constructing high resolution physical maps of genomic **DNA** which is useful for as a tool for isolating disease related genes. Current mapping methods are difficult and laborious. The new method combines the systematic quality of jumping and linking **libraries** with the convenience of the sequence tagged sites approach. (48pp)

L116 ANSWER 6 OF 22 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI
ACCESSION NUMBER: 2002-14180 BIOTECHDS
TITLE: Determining the relative abundance of **nucleic acid** sequences in different source populations, by hybridizing differentially labeled populations of DNAs from sources to be compared with reference **DNA** cloned on a support;
DNA hybridization, reference **DNA library**, microparticle support, vector expression in host cell, SNP detection and differentiation gene expression analysis
AUTHOR: FU R; **BRENNER S**; ALBRECHT G
PATENT ASSIGNEE: LYNX THERAPEUTICS INC
PATENT INFO: WO 2002027029 4 Apr 2002
APPLICATION INFO: WO 2000-US30396 27 Sep 2000
PRIORITY INFO: US 2000-235940 27 Sep 2000
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2002-416485 [44]
AB DERWENT ABSTRACT:
NOVELTY - Determining the relative abundance of a **nucleic acid** (NA) sequence among at least two NA populations, is new.
DETAILED DESCRIPTION - Determining (M) the relative abundance of a NA sequence among at least two NA populations, comprises: (a) contacting with a reference **library** which comprises multiple copies of a

Interestingly, sequences homologous to nodE, nodG, nodP, and nodQ have been assigned to another autonomously replicating element in Rhizobium species NGR234. Similarly one copy of the structural dctA gene is located on the symbiotic plasmid (dctA1) while the other is on what we assume to be the chromosome.

L116 ANSWER 3 OF 22 BIOTECHNO COPYRIGHT 2003 Elsevier Science B.V.
ACCESSION NUMBER: 1992:22179834 BIOTECHNO
TITLE: Encoded combinatorial chemistry
AUTHOR: **Brenner S.**; Lerner R.A.
CORPORATE SOURCE: Department of Chemistry, Scripps Research Institute, 10666 North Torrey Pines,La Jolla, CA 92037, United States.
SOURCE: Proceedings of the National Academy of Sciences of the United States of America, (1992), 89/12 (5381-5383)
CODEN: PNASA6 ISSN: 0027-8424
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
SUMMARY LANGUAGE: English

AB The diversity of chemical synthesis and the power of genetics are linked to provide a powerful, versatile method for drug screening. A process of alternating parallel combinatorial synthesis is used to encode individual members of a large **library** of chemicals with unique nucleotide sequences. After the chemical entity is bound to a target, the genetic tag can be amplified by replication and utilized for enrichment of the bound molecules by serial hybridization to a subset of the **library**. The nature of the chemical structure bound to the receptor is decoded by sequencing the nucleotide tag.

L116 ANSWER 4 OF 22 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI
ACCESSION NUMBER: 1999-12444 BIOTECHDS
TITLE: Analysis of differential gene expression by competitive hybridization to identify and isolate differentially expressed genes, e.g. for drug design;
production of differential expression **DNA library** for analysis by fluorescence-activated cell sorting, useful in drug design, pest control, therapeutics and diagnostics
AUTHOR: Albrecht G; **Brenner S**; Dubridge R
PATENT ASSIGNEE: Lynx-Ther.
LOCATION: Hayward, CA, USA.
PATENT INFO: WO 9935293 15 Jul 1999
APPLICATION INFO: WO 1999-US666 8 Jan 1999
PRIORITY INFO: US 1998-130446 6 Aug 1998; US 1998-5222 9 Jan 1998
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 1999-444205 [37]

AB A method of differential gene expression is claimed which comprises competitive hybridization of polynucleotide (I) populations of expressed genes from two different cell or tissue sources, with a reference population of sequences attached to separate solid phase supports in clonal subpopulations. Each (I) carries a light-generating label, a different one for each source. **Duplexes** between the expressed gene and reference population are formed at ratios directly proportional to the relative expression of the relevant gene in the two sources. Also claimed are: methods for determining the relative abundance's of gene products; a mixture of microparticles that carry many identical ss **nucleic acids** comprising an oligonucleotide tag attached to (I) from an mRNA of at least one cell or tissue source (useful for producing a differential expression **DNA library** which may be manipulated by fluorescence-activated cell sorting). The method is used for identifying and isolating

selected NA sequence, a probe (P1) derived from a first NA population, having a sequence complementary to the selected sequence and a terminal sample ID (SID) sequence and a probe (P2), derived from a second NA population, having a sequence complementary to the selected sequence and a terminal SID sequence, where P1 and P2 are present in relative amounts proportional to the abundance of the NA sequence in the respective populations, where upon contacting, P1 and P2 competitively hybridize with the selected sequence in the reference **library**, such that the probes are present in **duplexes** in relative amounts proportional to the abundance of the NA sequence in the respective populations, and the SID sequences are present as **single stranded** extensions on the **duplexes**, and the first SID sequences on the **duplexes** and the second SID sequences on the **duplexes** hybridize with each other in a 1:1 ratio; and (b) detecting the presence of unhybridized SID sequences, as an indication of the relative amounts of hybridized P1 and P2. INDEPENDENT CLAIMS are also included for the following: (1) a kit comprising a reference NA **library** containing a sequence present in several different sources, and P1 and P2; (2) a kit for use in preparation of SID tagged probes, competitive hybridization, SRQ decoding, for use in performing (M), comprising: (a) two or more SID adaptors for generating SID tagged probes, each adaptor comprising a double stranded oligonucleotide having in sequence: (i) a protruding **single strand** effective for ligation to a **DNA** restriction fragment (ii) an SID sequence; (iii) a restriction site; and (iv) a primer binding site, where cleavage by an enzyme recognizing the restriction site is effective to cleave all but elements (i) and (ii), from the adaptor, and the different adaptors have different SID sequences which are able to hybridize with each other; and (b) two or more SID decoders, selectively attachable to the different SID sequences with distinguishable light-generating labels.

BIOTECHNOLOGY - Preferred Method: Several probes derived from first and second populations are contacted with the reference **library** , and the reference **library** comprises multiple copies of the sequences present in the first and second populations, such that different sequences within the **library** are attached to spatially distinct solid phase supports in clonal subpopulations. The supports are separate regions of a planar support, or microparticles. Detection comprises attaching a labeled decoder group to unhybridized SID sequence. A light-generating label is present on decoder groups selectively attachable to unhybridized first SID sequences, and another distinguishable light-generating label is present on second decoder groups selectively attachable to unhybridized second SID sequences. Each decoder group includes a terminal oligonucleotide sequence that is complementary to either first or second SID sequence. The decoder labels are fluorescent dye molecules and each decoder label comprises multiple dye molecules. The spatially distinct solid phase supports are microparticles, and (M) also comprises sorting the microparticles by fluorescent activated cell sorting (FACS) according to the ratio of fluorescent signals generated by the fluorescent labels on each microparticle. (M) further comprises accumulating in a separate vessel, each microparticle having a value of the ratio of fluorescent signals within one or more selected ranges of values, and determining a nucleotide sequence of a portion of the NA sequence on one or more of the microparticles. P1 and P2 are further labeled with the first and second light-generating labels, respectively. A known fraction of P1 and P2 are further labeled with light-generating labels. The first and second SID sequences are complementary and hybridize with each other directly or through an intermediate molecule comprising sequences complementary to the SID sequences. (M) further comprises contacting the intermediate molecule with the reference **library** and P1 and P2.

USE - (M) is useful for determining the relative abundance of a NA sequence among two NA populations, and for analyzing differentially regulated or expressed genes, where the populations are cDNA

libraries derived from expressed genes of each of several sources such as different tissues, cells, individuals, and the reference **DNA library** is derived from genes expressed in the several different sources (M) is also useful for the analysis of genetic variations among individuals or population of individuals, where the genomic **DNA libraries** are derived from different individuals or populations of individuals, and the reference **DNA library** is derived from the pooled genomic **DNA** of such individuals or populations of individuals. (M) is also useful for sorting a population of NA sequences in accordance with their relative abundance between at least two NA populations (all claimed). (M) is useful for detecting the presence of differentially represented genetic variations including single nucleotide polymorphisms, deletions and duplications, in genomic **DNA** among different individuals.

ADVANTAGE - The genes whose expression levels change or are different than those of the other cells or tissues being examined may be analyzed separately from those that are not regulated or otherwise altered in response to stress or the condition being studied. **DNA** fragments representing genomic variations among individuals may be analyzed separately from those which are equally represented among the individual. The identity of the NA being analyzed, e.g. genomic **DNA**, cDNA, mRNA, RNA transcript, need not be known prior to analysis.

EXAMPLE - cDNA from each of the cell or tissue types of interest was prepared and directionally cloned into a vector. The mRNA extracted from such cells or tissues was combined, prior to first strand synthesis, and then a second strand synthesis was carried out. The resulting cDNAs were inserted into a vector containing a tag element (whose sequence is given in the specification). The vectors containing the tag-cDNA conjugates were then used to transform a suitable host, after which a sample of cells from the host culture was further expanded and vector **DNA** was extracted. The tag-cDNA conjugates were preferably amplified from the vectors by polymerase chain reaction (PCR) and processed for loading onto microparticles derivatized with tag complements. After the non-covalently attached strand was melted off, the cDNA-containing microparticle was ready to accept competitively hybridized gene products. A reference **DNA** population attached to microparticles was constructed from cDNA derived from stimulated THP-1 cells. Equal concentrations of labeled cDNAs from both stimulated (by treatment with phorbol 12-myristate 13-acetate (PMA) and lipopolysaccharide (LPS)) and unstimulated THP-1 cells were then competitively hybridized to the reference **DNA** population and the microparticles carrying the labeled cDNAs were analyzed by a fluorescent activated cell sorting (FACS) instrument, using non-SRQ probe ratio methods. THP-1 cells were grown in T-165 flasks containing Dulbecco's modified Eagle medium (DMEM)/F12 media (50 ml) supplemented with 10% fetal bovine serum, penicillin (100 units/mi), streptomycin (100 microg/ml) and beta-mercapto ethanol (0.5 muMI). Cells were treated with PMA and fresh media (without antibiotics) containing 10 microl of 5 mg/ml LPS in phosphate buffered saline (PBS) was added. Both the cultures of stimulated and unstimulated cells were incubated at 37 degrees C for 4 hours, after which cells were harvested. mRNA was extracted from 5x106 cells. A reference **DNA** population attached to microparticles were constructed from mRNA extracted from stimulated cells. Separate cDNA **libraries** were constructed from mRNA extracted from stimulated and unstimulated cells. 2.5 microg of rhodamine R110-labeled **single stranded DNA** was produced from the cDNA **library** derived from stimulated cells, and 2.5 microg of CY5-labeled **single stranded DNA** was produced from the cDNA **library** derived from unstimulated cells. The two 2.5 microg aliquots were mixed and competitively hybridized to the reference **DNA** on 9.34x105 microparticles. After hybridization, the microparticles were sorted by a Cytomation, Inc. MoFlo FACS instrument using non-SRQ labeled probes.

10000 microparticles corresponding to up-regulated genes were isolated, and 12000 microparticles corresponding to down-regulated genes were isolated. After melting off the labeled strands, the cDNAs carried by the microparticles were amplified and cloned into a cloning vector. After transformation, expansion of a host culture, and plating, 87 colonies of up-regulated cDNAs were picked and 73 colonies of down-regulated cDNAs were picked. The cDNAs were then sequenced. (88 pages)

L116 ANSWER 7 OF 22 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI
ACCESSION NUMBER: 2002-12460 BIOTECHDS
TITLE: Generating reference **library** of restriction fragments from pooled **nucleic acids**, by using reference population of restriction fragments to compare frequencies of polymorphic sequences between different population pools;
restriction fragment reference **library** generation, **single stranded** nuclease, and polymerase chain reaction for disease-associated gene identification and plant-associated phenotype trait screening
AUTHOR: **BRENNER S**
PATENT ASSIGNEE: LYNX THERAPEUTICS INC
PATENT INFO: WO 2002016645 28 Feb 2002
APPLICATION INFO: WO 2000-US26115 21 Aug 2000
PRIORITY INFO: US 2000-227058 21 Aug 2000
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2002-315468 [35]

N No report

AB DERWENT ABSTRACT:

NOVELTY - Generating (M1) a reference **library** comprising a mixture of **heterologous nucleic acid** fragments, is new.

DETAILED DESCRIPTION - Generating a reference **library** comprising a mixture of **heterologous nucleic acid** fragments, comprising: (1) digesting pooled **nucleic acids** comprising first restriction site with a first RE to produce a mixture of RF; (2) forming first population (P1) of ssDNA fragments from a first subpopulation of RF, which comprise a second restriction site which is different from the first restriction site; (3) forming second population (P2) of ssDNA fragments from a second subpopulation of RF, which do not contain second restriction site, and where the first ssDNA fragments have complementary sequences to the second ssDNA fragments from the second subpopulation when the ssDNA fragments are derived from the same RF; (4) hybridizing P1 and P2 of ssDNA fragments to form a first population of **duplexes**; and (5) treating the first population of **duplexes** with a **single strand** dependent nuclease to digest mismatched **duplexes** in the first population.

BIOTECHNOLOGY - Preferred Method: The method further includes reannealing intact ssDNA from the nuclease digestion to form a second population of **duplexes**, isolating the second population of **duplexes** using biotin precipitation to form a reference population of RF, and amplifying the matched **duplexes** using polymerase chain reaction (PCR). The **single strand** dependent nuclease is S1, mungbean **endonuclease**, especially T7 **endonuclease**. The method involves digesting pooled **nucleic acid** with RE to produce a mixture of F1 having first cleavage ends and ligating an **Exo III** resistant linker to the first cleavage ends of F1 to form a first ligation product (LP1). LP1 is digested with a second RE to form a mixture of second RF (F2), some of which comprise a second cleavage end, and is ligated to an **Exo III** susceptible linker to form a second ligation product (LP2) which includes LP1. The linker comprises a first member of

a binding pair. LP2 is digested with **Exo III** to form a third ligation product (LP3) population comprising ssDNA comprising end sequences corresponding to the **Exo III** resistant and **Exo III** susceptible linkers, and double stranded **DNA** comprising end sequences corresponding to the **Exo III** resistant linkers. LP3 is denatured and hybridized to form a reannealed LP3 population. The annealed LP3 population is contacted with a second member of the binding pair to enrich for **duplexes** which form a reference population of RF and is further contacted with exonuclease I. The **Exo III** susceptible linker further comprises biotin.

USE - The method is useful for making a reference **library** of RF from pooled **nucleic acids** the contain a sequence polymorphism. The reference **libraries** are **heterogeneous** mixtures enriched for polymorphic **nucleic acid** fragments. Such **libraries** are useful in identifying single or multiple alleles which are associated with different phenotypes, and to compare the frequency of various polymorphisms in a population of interest. Polymorphisms which occur more frequently in one population than another, can be isolated and identified. When used to analyze other populations, a pool of **DNA** from individuals having a first phenotype is compared to a population which demonstrates a second phenotype. The reference **libraries** can be used to screen for polymorphic markers in close proximity to genes which may be associated with one or more phenotypes or genotypes. Polymorphisms associated with genotypes which show simple Mendelian inheritance, as well as genotypes or phenotypes associated with a complex trait can be detected. Other phenotypes of biological interest which can be screened include common diseases in humans such as cardiovascular diseases, autoimmune diseases, cancer, diabetes, schizophrenia, bipolar disorder and other psychiatric disorders. In addition, polymorphisms in other organisms, i.e., plants, associated with phenotypical traits such as disease resistance and yield can also be screened. The polymorphic probes from the reference **library** are useful to compare the frequency of various polymorphisms between different pools of **nucleic acids**.

ADVANTAGE - The method provides a significant improvement over conventional marker associated studies, as no sequence information is required to generate and use the reference **libraries**.

EXAMPLE - A conventional pUC19 plasmid was modified to create two additional Sau 3A sites between the Taq I sites located at base positions 430 and 906 of the plasmid. This newly created plasmid (p0T2S) was then modified further with the addition of a Taq I site between the two new Sau 3A sites, to create the plasmid p1T2S. The two plasmids were polymorphic at the new Taq 1 site. The two plasmids were digested separately with Sau 3A. **Single stranded** portions of Sau 3A fragments containing Taq I sites were generated using adaptors and primers given in the specification. The Sau 3A digested p1T2S plasmid was filled in with dGTP and then excess of Q adaptors was added in a conventional ligation reaction to form product, which was then digested with Taq I to give three possible products. To this mixture, an excess of M adaptors were added in a conventional ligation reaction to form the three possible products (A), (B), and (C). After ligating M adaptors, the mixture was treated with **exonuclease III** to render fragments (A) and (B) **single stranded**. M and Q primers were then added to the reaction mixture and polymerase chain reaction (PCR) was carried out to form product which was then digested with Sau 3A to remove the Q adaptor. The resulting fragment was then treated with T7 gene 6 5'-exonuclease to produce **single stranded** fragments (F1). **Single stranded** portions of Sau 3A fragments lacking Taq I sites (Taq fragments) were generated from the plasmid p0T2S. The Sau 3A digested p0T2S was filled in with dGTP and then an excess of N adaptors were added in a conventional

ligation reaction to form product, which was then digested with Taq I to give three possible products (D), (E), and (F). The reaction mixture was then treated with T7 gene 6 exonuclease to render all fragments **single stranded**, except those (D) having two N adaptors attached. After treatment with exonuclease I to eliminate **single stranded** fragments, N primers were added to the reaction mixture and PCR was carried out to enrich the mixture for fragment (D). The resulting fragments were then treated with **exonuclease III** to produce **single stranded** fragments (F2). Fragments (F1) and (F2) were annealed and the 3' strands of the resulting **duplexes** were extended with T4 **DNA** polymerase to form fragments having primer binding sites for M and N primers. M and N primers were added to the reaction mixture and the fragments were copied by PCR. The PCR amplicons from the reaction were separated by gel electrophoresis and two fragments were identified.(82 pages)

L116 ANSWER 8 OF 22 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI
ACCESSION NUMBER: 2000-13522 BIOTECHDS
TITLE: **Nucleic acid** reference **library** containing a **heterogeneous** mixture of **nucleic acid** fragments which contain a portion of a polymorphic subregion of a polymorphic consensus sequence, useful for genetic identification and comparison; **DNA** probe array
AUTHOR: **Brenner S**
PATENT ASSIGNEE: Lynx-Ther.
LOCATION: Hayward, CA, USA.
PATENT INFO: WO 2000050632 31 Aug 2000
APPLICATION INFO: WO 2000-US4349 18 Feb 2000
PRIORITY INFO: US 1999-158483 8 Oct 1999; Us 1999-121023 22 Feb 1999
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2000-533184 [48]

r ufound ~~internatio~~ report

AB A **nucleic acid** reference **library** containing a **heterogeneous** mixture of **nucleic acid** fragments which contain a portion of a polymorphic subregion of a polymorphic consensus sequences, where each of the polymorphic subregions is bound by first restriction sites and contains an internal polymorphic restriction site different from the first site, is new. Also claimed are: an array containing a solid support having defined regions on the surface, where each region contains a different polymorphic **DNA** probe and where each of polymorphic **DNA** probes contain a portion of polymorphic consensus sequence; a **nucleic acid** reference **library** of genomic **DNA**; a method for making a reference **library** containing a mixture of **heterogeneous nucleic acid** fragments; and a method for determining the ratio of a polymorphic subregion between at least 2 different pools of test **nucleic acid**. The **library** is useful for rapid and sensitive genome-wide identification of differences in genetic composition between groups of individuals, particularly when screening for diseases. (91pp)

L116 ANSWER 9 OF 22 CAPLUS COPYRIGHT 2003 ACS DUPLICATE 1
ACCESSION NUMBER: 2002:256515 CAPLUS
DOCUMENT NUMBER: 136:274222
TITLE: Method for determining relative abundance of nucleic acid sequences by source-specific tagging and competitive hybridization
INVENTOR(S): Fu, Rongdian; **Brenner, Sydney**; Albrecht, Glenn
PATENT ASSIGNEE(S): Lynx Therapeutics, Inc., USA
SOURCE: PCT Int. Appl., 88 pp.

```
                       CODEN: PIXXD2
DOCUMENT TYPE:         Patent
LANGUAGE:              English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

    PATENT NO.        KIND  DATE        APPLICATION NO.   DATE
    ---------------   ----  --------    ---------------   --------
    WO 2002027029      A2   20020404    WO 2001-US30396   20010927
        W:  AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN,
            CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH,
            GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR,
            LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PH, PL,
            PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG,
            UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
        RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY,
            DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF,
            BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG
    AU 2001093163      A5   20020408    AU 2001-93163     20010927
    US 2003027157      A1   20030206    US 2001-967238    20010927
PRIORITY APPLN. INFO.:               US 2000-235940P  P  20000927
                                     WO 2001-US30396  W  20010927
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AB Disclosed are methods for identifying nucleic acid sequences which are of different abundances in different nucleic acid source populations, e.g. differentially expressed genes or genomic variations among individuals or populations of individuals. In one embodiment, probes derived from the source nucleic acid populations are derivatized with a terminal sample ID (SID) sequence characteristic of that population. Upon competitive hybridization of the probes to a ref. or index nucleic acid library contg. all the sequences in the populations being compared, the SID tags remain single stranded, and those from the different sources are then annealed to one another. Unhybridized (remainder) SID sequences are then quantified. By labeling such remainder SID sequences with a fluorescent dye, FACS sorting of beads contg. the hybridized probes can be carried out. The signal ratio upon which such sorting is based is enhanced compared to competitive hybridization using labeled probes without SID sequences.

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L116 ANSWER 10 OF 22  CAPLUS  COPYRIGHT 2003 ACS      DUPLICATE 2
ACCESSION NUMBER:      2002:158030  CAPLUS
DOCUMENT NUMBER:       136:211839
TITLE:                 Reference library comprising polymorphic DNA
                       fragments and uses in genetic identification and
                       comparison
INVENTOR(S):           Brenner, Sydney
PATENT ASSIGNEE(S):    Lynx Therapeutics, Inc., USA
SOURCE:                PCT Int. Appl., 82 pp.
                       CODEN: PIXXD2
DOCUMENT TYPE:         Patent
LANGUAGE:              English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

    PATENT NO.        KIND  DATE        APPLICATION NO.   DATE
    ---------------   ----  --------    ---------------   --------
    WO 2002016645      A2   20020228    WO 2001-US26115   20010821
        W:  AU, CA, JP
        RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL,
            PT, SE, TR
    AU 2001085155      A5   20020304    AU 2001-85155     20010821
    US 2003032020      A1   20030213    US 2001-934020    20010821
PRIORITY APPLN. INFO.:               US 2000-227058P  P  20000821
                                     WO 2001-US26115  W  20010821
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a duplicate of #7

AB The invention provides methods and materials for generating a ref. library

of restriction fragments from pooled nucleic acids that contain a sequence polymorphism. An important aspect of the invention is the use of the ref. population of restriction fragments to compare the frequencies of polymorphic sequences between different populations pools. Such comparisons may be accomplished by competitively hybridizing DNA from the resp. pools which has been enriched for the presence of a restriction site polymorphism with DNA from the ref. population. Preferably, such competitive hybridization reactions are carried out on the ref. library attached to one or more solid phase supports. Most preferably, members of the ref. library are attached to individual microparticle so that each microparticle has a unique fragment attached. After competitive hybridization, the microparticles may be analyzed and sorted to identify those microparticles carrying sequences for which the pools being compared exhibit different polymorphic frequencies.

L116 ANSWER 11 OF 22 CAPLUS COPYRIGHT 2003 ACS DUPLICATE 3
ACCESSION NUMBER: 2000:608938 CAPLUS
DOCUMENT NUMBER: 133:203823
TITLE: Preparation of genomic **libraries** enriched in sequences showing restriction site polymorphisms
INVENTOR(S): **Brenner, Sydney**
PATENT ASSIGNEE(S): Lynx Therapeutics, Inc., USA
SOURCE: PCT Int. Appl., 93 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

✓ duplicate of #8

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2000050632	A2	20000831	WO 2000-US4349	20000218
WO 2000050632	A3	20010329		

W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

EP 1157131	A2	20011128	EP 2000-910255	20000218

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO

JP 2002537774	T2	20021112	JP 2000-601195	20000218

PRIORITY APPLN. INFO.:
US 1999-121023P P 19990222
US 1999-158483P P 19991008
WO 2000-US4349 W 20000218

AB The invention provides methods and materials for generating a ref. library of restriction fragments from pooled nucleic acids that contain a sequence polymorphism. Preferably, such a library is formed by digesting genomic DNA from a pool of individuals with a first and a second restriction endonuclease to form a population of restriction fragments; isolating restriction fragments of the population digested by both the first and second restriction endonucleases and forming a first single stranded fragment population therefrom; sep. isolating restriction fragments from the population digested by the first restriction endonuclease but not the second restriction endonuclease and forming a second single stranded fragment population therefrom; hybridizing the first and second single stranded fragment populations to form a population of duplexes; and isolating the population of duplexes to form a ref. library of restriction fragments that contain sequence polymorphism. An important aspect of the

invention is the use of the ref. population of restriction fragments to compare the frequencies of polymorphic sequences between different population pools. Such comparisons may be accomplished by competitively hybridizing DNA from the resp. pools which has been enriched for the presence of a restriction site polymorphism with DNA from the ref. population. Preferably, such competitive hybridization reactions are carried out the ref. library attached to one or more solid phase supports. Most preferably, members of the ref. library are attached to individual microparticles so that each microparticle has a unique fragment attached. After competitive hybridization, the microparticles may be analyzed and sorted for identifying those microparticles carrying sequences for which the pools being compared exhibit different polymorphic frequencies.

L116 ANSWER 12 OF 22 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2003:71751 CAPLUS
DOCUMENT NUMBER: 138:101955
TITLE: Differentially expressed genes selected by optical sorting of cDNA **libraries** immobilized on microparticles and competitively hybridized to labeled probes
INVENTOR(S): Albrecht, Glenn; **Brenner, Sydney**; Dubridge, Robert B.
PATENT ASSIGNEE(S): Lynx Therapeutics, Inc., USA
SOURCE: U.S., 48 pp., Cont.-in-part of U.S. 6,265,163. CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 3
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 6511802	B1	20030128	US 1999-227694	19990108
US 6265163	B1	20010724	US 1998-130546	19980806

PRIORITY APPLN. INFO.: US 1998-5222 B2 19980109
US 1998-130546 A2 19980806

AB The invention provides a method and materials for monitoring and isolating differentially expressed genes. In accordance with the method of the invention, differently labeled populations of DNAs from sources to be compared are competitively hybridized with ref. DNA cloned on solid phase supports, e.g. microparticles, to provide a differential expression library which, in the preferred embodiment, may be manipulated by fluorescence-activated cell sorting (FACS). Monitoring the relative signal intensity of the different fluorescent labels on the microparticles permits quant. anal. of expression levels relative to the ref. DNA. Populations of microparticles having relative signal intensities of interest can be isolated by FACS and the attached DNAs identified by sequencing, such as with massively parallel signature sequencing (MPSS), or with conventional DNA sequencing protocols. Using these techniques, differential gene expression was analyzed in THP-1 cell line and in Saccharomyces cerevisiae.

REFERENCE COUNT: 27 THERE ARE 27 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L116 ANSWER 13 OF 22 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2002:461223 CAPLUS
DOCUMENT NUMBER: 137:42548
TITLE: Planar arrays of microparticle-bound polynucleotides
INVENTOR(S): Bridgham, John; Corcoran, Kevin; Golda, George; Pallas, Michael C.; **Brenner, Sydney**
PATENT ASSIGNEE(S): Lynx Therapeutics, Inc., USA
SOURCE: U.S., 25 pp., Cont.-in-part of U.S. Ser. No. 862,610, abandoned.

CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 10
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 6406848	B1	20020618	US 1999-424028	19991116
EP 931165	A1	19990728	EP 1996-940238	19961011
R: CH, DE, FR, GB, LI				
JP 2000511045	T2	20000829	JP 1997-515240	19961011
WO 9853300	A2	19981126	WO 1998-US11224	19980522
WO 9853300	A3	19990225		
W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, GW, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
US 6228589	B1	20010508	US 2000-269911	20000228
US 2002051992	A1	20020502	US 2001-908130	20010717
US 2002061529	A1	20020523	US 2001-908131	20010717
US 2002137052	A1	20020926	US 2001-907795	20010717

PRIORITY APPLN. INFO.:
US 1997-862610 B2 19970523
WO 1998-US11224 W 19980522
WO 1996-US16342 W 19961011
US 1999-424028 A3 19991116

AB An app. and system are provided for simultaneously analyzing a plurality of analytes anchored to microparticles. Microparticles each having a uniform population of a single kind of analyte attached are disposed as a substantially immobilized planar array inside of a flow chamber where steps of an anal. process are carried out by delivering a sequence of processing reagents to the microparticles by a fluidic system under microprocessor control. In response to such process steps, an optical signal is generated at the surface of each microparticle which is characteristic of the interaction between the analyte carried by the microparticle and the delivered processing reagent. The plurality of analytes are simultaneously analyzed by collecting and recording images of the optical signals generated by all the microparticles in the planar array. A key feature of the invention is the correlation of the sequence of optical signals generated by each microparticle in the planar array during the anal. process.

REFERENCE COUNT: 2 THERE ARE 2 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L116 ANSWER 14 OF 22 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2001:537423 CAPLUS
DOCUMENT NUMBER: 135:133102
TITLE: Solid phase selection of differentially expressed genes with limited sequence information
INVENTOR(S): Albrecht, Glenn; **Brenner, Sydney**; Dubridge, Robert B.
PATENT ASSIGNEE(S): Lynx Therapeutics, Inc., USA
SOURCE: U.S., 33 pp., Cont.-in-part of U.S. Ser. No. 5,222, abandoned.
CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 3
PATENT INFORMATION:

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PATENT NO.       KIND  DATE          APPLICATION NO.  DATE
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US 6265163        B1   20010724      US 1998-130546   19980806
CA 2317695        AA   19990715      CA 1999-2317695  19990108
WO 9935293        A2   19990715      WO 1999-US666    19990108
WO 9935293        A3   19990930
    W:  AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE,
        DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP,
        KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN,
        MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM,
        TR, TT, UA, UG, US, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU,
        TJ, TM
    RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES,
        FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI,
        CM, GA, GN, GW, ML, MR, NE, SN, TD, TG
AU 9921139        A1   19990726      AU 1999-21139    19990108
AU 754929         B2   20021128
EP 1054999        A2   20001129      EP 1999-901448   19990108
    R:  AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT,
        IE, FI
JP 2002500050     T2   20020108      JP 2000-527674   19990108
US 6511802        B1   20030128      US 1999-227694   19990108
NO 2000003531     A    20000905      NO 2000-3531     20000707
PRIORITY APPLN. INFO.:            US 1998-5222     B2 19980109
                                  US 1998-130546   A  19980806
                                  WO 1999-US666    W  19990108
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AB The invention provides a method and materials for monitoring and isolating differentially expressed genes. In accordance with the method of the invention, differently labeled populations of DNAs from sources to be compared are competitively hybridized with ref. DNA cloned on solid phase supports, e.g. microparticles, to provide a differential expression library which, in the preferred embodiment, may be manipulated by fluorescence-activated cell sorting (FACS). Monitoring the relative signal intensity of the different fluoroscent labels on the microparticles permits quant. anal. of expression levels relative to the ref. DNA. Populations of microparticles having relative signal intensities of interest can be isolated by FACS and the attached DNAs identified by sequencing, such as with massively parallel signature sequencing (MPSS), or with conventional DNA sequencing protocols. Detailed exptl. protocols for the prepn. of probe libraries are given.

REFERENCE COUNT: 70 THERE ARE 70 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L116 ANSWER 15 OF 22 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2000:154838 CAPLUS
DOCUMENT NUMBER: 132:203759
TITLE: In vitro cloning of complex mixtures of DNA on microbeads: physical separation of differentially expressed cDNAs
AUTHOR(S): **Brenner, Sydney**; Williams, Steven R.; Vermaas, Eric H.; Storck, Thorsten; Moon, Keith; McCollum, Christie; Mao, Jen-I.; Luo, Shujun; Kirchner, James J.; Eletr, Sam; DuBridge, Robert B.; Burcham, Timothy; Albrecht, Glenn
CORPORATE SOURCE: Lynx Therapeutics, Inc., Hayward, CA, 94545, USA
SOURCE: Proceedings of the National Academy of Sciences of the United States of America (2000), 97(4), 1665-1670
CODEN: PNASA6; ISSN: 0027-8424
PUBLISHER: National Academy of Sciences
DOCUMENT TYPE: Journal
LANGUAGE: English
AB A method for cloning nucleic acid mols. onto the surfaces of 5-.mu.m

microbeads rather than in biol. hosts is described. A unique tag sequence is attached to each mol., and the tagged library is amplified. Unique tagging of the mols. is achieved by sampling a small fraction (1%) of a very large repertoire of tag sequences. The resulting library is hybridized to microbeads that each carry .apprx.106 strands complementary to one of the tags. About 105 copies of each mol. are collected on each microbead. Because such clones are segregated on microbeads, they can be operated on simultaneously and then assayed sep. The utility of this approach is demonstrated by labeling and extg. microbeads bearing clones differentially expressed between 2 libraries by using a fluorescence-activated cell sorter (FACS). Because no prior information about the cloned mols. is required, this process is obviously useful where sequence databases are incomplete or nonexistent. More importantly, the process also permits the isolation of clones that are expressed only in given tissues or that are differentially expressed between normal and diseased states. Such clones then may be spotted on much more cost-effective, tissue-or disease-directed, low-d. planar microarrays.

REFERENCE COUNT: 14 THERE ARE 14 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L116 ANSWER 16 OF 22 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1999:633269 CAPLUS
DOCUMENT NUMBER: 131:267933
TITLE: A method for sequencing very long DNAs with a small set of primers that can be mutated and adapted to novel sequence information
INVENTOR(S): **Brenner, Sydney**
PATENT ASSIGNEE(S): Lynx Therapeutics, Inc., USA
SOURCE: U.S., 28 pp., Cont.-in-part of U.S. 5,780,231. CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 4
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 5962228	A	19991005	US 1997-916120	19970822
US 5763175	A	19980609	US 1995-560313	19951117
US 5780231	A	19980714	US 1996-611155	19960305
JP 11151092	A2	19990608	JP 1998-237840	19980824

PRIORITY APPLN. INFO.: US 1995-560313 19951117
US 1996-611155 19960305
US 1997-916120 19970822

AB A novel "primer walking" method for DNA sequencing is provided that uses repeated cycles of nucleotide identification by selective extension and primer advancement along a template by template mutation. An important feature of the invention is providing a set of primers, referred to herein as "rolling primers" that contain complexity-reducing nucleotides for reducing the no. of primers required for annealing to every possible primer binding site on a sequencing template. These primers have a defined 3'-terminal region contg. complexity-reducing nucleotides (i.e. bases showing ambiguous base pairing, such as 2'-deoxyinosine), and a 5'-region contg. a unique sequence tag that allows it to be captured by a complementary sequence that is part of an ordered array. Another important feature of the invention is the systematic replacement of at least one of the four nucleotides in the target polynucleotide with its cognate complexity-reducing nucleotide or complement thereof. Sequencing is initiated by annealing rolling primers differing only in their terminal nucleotides to a primer binding site of a sequencing template so that only the rolling primer whose terminal nucleotide forms a perfect complement with the template leads to the formation of an extension product. After amplifying the double stranded extension product to form an amplicon, the

terminal nucleotide, and hence its complement in the template, is identified by the identity of the amplicon. The primer binding site of the template of the successfully amplified polynucleotide is then mutated by, for example, oligonucleotide-directed mutagenesis so that a subsequent rolling primer may be selected from the set that forms a perfectly matched duplex with the mutated template at a site which is shifted towards the direction of extension by one nucleotide relative to the binding site of the previous rolling primer. The steps of selective extension, amplification and identification are then repeated. In this manner, the primers "roll" along the polynucleotide during the sequencing process, moving a base at a time along the template with each cycle. The procedure may be readily automated for large-scale sequencing projects. Use of inosine as a base in combination with other bases in the 4 3'-terminal bases allows a set of of six primers to act as the progenitors of 200 or more primers that can be generated by mutagenesis as needed.

REFERENCE COUNT: 8 THERE ARE 8 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L116 ANSWER 17 OF 22 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 1998:790734 CAPLUS
DOCUMENT NUMBER: 130:21343
TITLE: System and apparatus for sequential processing of analytes
INVENTOR(S): Pallas, Michael C.; **Brenner, Sydney**; Bridgham, John; Corcoran, Kevin; Golda, George
PATENT ASSIGNEE(S): Lynx Therapeutics, Inc., USA
SOURCE: PCT Int. Appl., 43 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 10
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9853300	A2	19981126	WO 1998-US11224	19980522
WO 9853300	A3	19990225		
W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, GW, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
EP 931165	A1	19990728	EP 1996-940238	19961011
R: CH, DE, FR, GB, LI				
JP 2000511045	T2	20000829	JP 1997-515240	19961011
AU 9877155	A1	19981211	AU 1998-77155	19980522
AU 736321	B2	20010726		
EP 985142	A2	20000315	EP 1998-925137	19980522
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO				
JP 2002507280	T2	20020305	JP 1998-550757	19980522
US 6406848	B1	20020618	US 1999-424028	19991116
US 6228589	B1	20010508	US 2000-269911	20000228
US 2002051992	A1	20020502	US 2001-908130	20010717
US 2002061529	A1	20020523	US 2001-908131	20010717
US 2002137052	A1	20020926	US 2001-907795	20010717

PRIORITY APPLN. INFO.:

US 1997-862610	A	19970523
WO 1996-US16342	W	19961011
WO 1998-US11224	W	19980522
US 1999-424028	A3	19991116

AB An app. and system are provided for simultaneously analyzing a plurality of analytes anchored to microparticles. Microparticles each having a uniform population of a single kind of analyte attached are disposed as a substantially immobilized planar array inside of a flow chamber where steps of an anal. process are carried out by delivering a sequence of processing reagents to the microparticles by a fluidic system under microprocessor control. In response to such process steps, an optical signal is generated at the surface of each microparticle which is characteristic of the interaction between the analyte carried by the microparticle and the delivered processing reagent. The plurality of analytes are simultaneously analyzed by collecting and recording images of the optical signals generated by all the microparticles in the planar array. A key feature of the invention is the correlation of the sequence of optical signals generated by each microparticle in the planar array during the anal. process.

L116 ANSWER 18 OF 22 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1998:803923 CAPLUS
DOCUMENT NUMBER: 130:48286
TITLE: oligonucleotide probe sets with members that do not cross-hybridize for sorting and identification of nucleic acid sequences
INVENTOR(S): **Brenner, Sydney**; Albrecht, Glenn; Macevicz, Stephen C.
PATENT ASSIGNEE(S): Lynx Therapeutics, Inc., USA
SOURCE: U.S., 38 pp., Cont.-in-part of U.S. 5,604,097. CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 10
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 5846719	A	19981208	US 1996-659453	19960606
US 5604097	A	19970218	US 1994-358810	19941219
EP 931165	A1	19990728	EP 1996-940238	19961011
R: CH, DE, FR, GB, LI				
JP 2000511045	T2	20000829	JP 1997-515240	19961011
WO 9746704	A1	19971211	WO 1997-US9472	19970602
W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, GH, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, KE, LS, MW, SD, SZ, UG, AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
AU 9733740	A1	19980105	AU 1997-33740	19970602
AU 733782	B2	20010524		
EP 923650	A1	19990623	EP 1997-929757	19970602
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
CN 1230226	A	19990929	CN 1997-197101	19970602
JP 2000515006	T2	20001114	JP 1998-500755	19970602
US 6013445	A	20000111	US 1997-946138	19971007
US 6352828	B1	20020305	US 1998-53116	19980401
US 6138077	A	20001024	US 1998-89853	19980603
US 6280935	B1	20010828	US 1998-90809	19980604
US 6172218	B1	20010109	US 1998-92226	19980605
US 6172214	B1	20010109	US 1998-131009	19980807
US 6235475	B1	20010522	US 1998-130862	19980807
US 6150516	A	20001121	US 1998-196543	19981120

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
NO 9805698	A	19990208	NO 1998-5698	19981204
US 6228589	B1	20010508	US 2000-269911	20000228

PRIORITY APPLN. INFO.:
US 1994-322348 B2 19941013
US 1994-358810 A2 19941219
WO 1995-US12791 A 19951012
US 1996-659453 A 19960606
US 1996-689587 A 19960812
WO 1996-US16342 W 19961011
US 1997-862610 B2 19970523
WO 1997-US9472 W 19970602

AB The invention provides a method of tracking, identifying, and/or sorting classes or subpopulations of mols. by the use of oligonucleotide tags. Oligonucleotide tags of the invention comprise oligonucleotides selected from a minimally cross-hybridizing set. Preferably, such oligonucleotides each consist of a plurality of subunits 3 to 9 nucleotides in length. A subunit of a minimally cross-hybridizing set forms a duplex or triplex having two or more mismatches with the complement of any other subunit of the same set. The no. of oligonucleotide tags available in a particular embodiment depends on the no. of subunits per tag and on the length of the subunit. The tag set may contain 1,000-500,000 members. An important aspect of the invention is the use of the oligonucleotide tags for sorting polynucleotides by specifically hybridizing tags attached to the polynucleotides to their complements on solid phase supports. This embodiment provides a readily automated system for manipulating and sorting polynucleotides, particularly useful in large-scale parallel operations, such as large-scale DNA sequencing, mRNA fingerprinting, and the like, wherein many target polynucleotides or many segments of a single target polynucleotide are sequenced simultaneously.

REFERENCE COUNT: 18 THERE ARE 18 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L116 ANSWER 19 OF 22 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1997:324743 CAPLUS
DOCUMENT NUMBER: 127:49307
TITLE: Lambdoid bacteriophage vectors for expression and display of foreign proteins as fusion products with phage tail protein matrix-anchoring domain
INVENTOR(S): Maruyama, Ichiro; Maruyama, Hiroko; **Brenner, Sydney**
PATENT ASSIGNEE(S): Scripps Research Institute, USA
SOURCE: U.S., 41 pp.
CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 5627024	A	19970506	US 1994-286888	19940805
US 2002110802	A1	20020815	US 1997-852020	19970506

PRIORITY APPLN. INFO.: US 1994-286888 A3 19940805

AB Lambdoid bacteriophage vectors are disclosed which comprise a matrix of proteins encapsulating a genome. The genome encodes autogenously assembling protein subunits surface-integrated into the matrix via a lambdoid phage tail protein matrix-anchoring domain fused to at least one of the subunits. The .lambda.foo vector is provided in one example and phage library display is included.

L116 ANSWER 20 OF 22 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER: 1993:58037 BIOSIS
DOCUMENT NUMBER: PREV199395034339
TITLE: A selective lambda phage cloning vector with automatic

excision of the insert in a plasmid.
AUTHOR(S): Maruyama, Ichiro N. (1); **Brenner, Sydney**
CORPORATE SOURCE: (1) Dep. Cell Biology, Scripps Res. Inst., 10666 North Torrey Pines Road, La Jolla, Calif. 92037 USA
SOURCE: Gene (Amsterdam), (1992) Vol. 120, No. 2, pp. 135-141. ISSN: 0378-1119.
DOCUMENT TYPE: Article
LANGUAGE: English
AB A bacteriophage lambda cloning vehicle has been constructed for the generation of cDNA **libraries**. The vector has the following properties. (1) It has a unique BamHI site engineered into the lambda gam gene. Segments of **DNA** can be cloned into this site and clones with an insert can be selected by their ability to grow on an Escherichia coli host lysogenic for phage P2 (Spi- phenotype). (2) When the recombinant phage infects a Cre-producing E. coli strain, a site-specific recombination event results in the excision of a plasmid replicon with the cloned insert. (3) **Single-stranded** DNAs can be recovered by growing helper M13 phages on bacteria harboring such plasmids. The vector, lambda-MGU2, has been used to construct a nematode (Caenorhabditis elegans) cDNA **library**.

L116 ANSWER 21 OF 22 WPIDS (C) 2003 THOMSON DERWENT
ACCESSION NUMBER: 1997-099943 [09] WPIDS
CROSS REFERENCE: 1996-222001 [22]; 1996-222023 [22]; 1998-042210 [04]; 1999-024716 [02]; 2000-170257 [14]
DOC. NO. CPI: C1997-031923
TITLE: Sorting poly nucleotide(s) on to solid supports by attachment to oligo nucleotide tags - then specific hybridisation of tags to immobilised complement, e.g. for automated **DNA** mapping and sequencing, genetic identification and diagnosis.
DERWENT CLASS: B04 D16
INVENTOR(S): ALBRECHT, G; **BRENNER, S**
PATENT ASSIGNEE(S): (LYNX-N) LYNX THERAPEUTICS INC; (SPEC-N) SPECTRAGEN INC
COUNTRY COUNT: 36
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 9641011	A1	19961219	(199709)*	EN	79
RW: AT BE CH DE DK ES FI FR GB GR IE IT LU MC NL PT SE					
W: AU BR CA CN CZ EE FI HU JP KR LT LV NO NZ PL RU SG SI SK					
AU 9661020	A	19961230	(199716)		
NO 9705744	A	19980205	(199816)		
EP 832287	A1	19980401	(199817)	EN	
R: AT BE CH DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE					
CZ 9703926	A3	19980617	(199830)		
CN 1193357	A	19980916	(199905)		
HU 9900910	A2	19990728	(199936)		
JP 11507528	W	19990706	(199937)		91
KR 99022543	A	19990325	(200023)		
AU 718357	B	20000413	(200028)		

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 9641011	A1	WO 1996-US9513	19960606
AU 9661020	A	AU 1996-61020	19960606
NO 9705744	A	WO 1996-US9513	19960606
		NO 1997-5744	19971205
EP 832287	A1	EP 1996-918333	19960606
		WO 1996-US9513	19960606

CZ 9703926	A3	WO 1996-US9513	19960606
		CZ 1997-3926	19960606
CN 1193357	A	CN 1996-196135	19960606
HU 9900910	A2	WO 1996-US9513	19960606
		HU 1999-910	19960606
JP 11507528	W	WO 1996-US9513	19960606
		JP 1997-501818	19960606
KR 99022543	A	WO 1996-US9513	19960606
		KR 1997-709024	19971206
AU 718357	B	AU 1996-61020	19960606

FILING DETAILS:

PATENT NO	KIND		PATENT NO
AU 9661020	A	Based on	WO 9641011
EP 832287	A1	Based on	WO 9641011
CZ 9703926	A3	Based on	WO 9641011
HU 9900910	A2	Based on	WO 9641011
JP 11507528	W	Based on	WO 9641011
KR 99022543	A	Based on	WO 9641011
AU 718357	B	Previous Publ.	AU 9661020
		Based on	WO 9641011

PRIORITY APPLN. INFO: WO 1995-US12791 19951012; US 1995-478238 19950607

AB WO 9641011 A UPAB: 20001230

Components of a population of polynucleotides (I) are sorted on to 1 or more solid supports by: (a) attaching an oligonucleotide tag (from a repertoire) to each (I), each tag being minimally cross-hybridising; (b) sampling the population so that all different (I) have different tags; (c) sorting by specific hybridisation of the tags with their complements which are attached as uniform populations of identical oligonucleotides in spatially distinct regions on one or more solid phase support.

USE - The method is used (a) to identify a population of mRNA molecules; (b) to detect presence/absence of selected target sequences in a target (I); or (c) to identify (I), including new ones in cDNA **libraries**, e.g. for construction and use of combinatorial chemical **libraries**; large scale **DNA** mapping and sequencing; genetic identification; medical diagnosis (e.g. analysis of gene expression in diseased and normal tissue) etc.

ADVANTAGE - The method is easily automated for manipulation and sorting of (I) in large scale parallel processing where many target (I), or many target segments of a single (I), are sequenced simultaneously. The no. of separate template prepn. steps required for sequencing is reduced. Since the stability of any mismatch duplex or triplex between a tag and complement of a different tag is so much less than that of the complex between the tag and its exact complement, problems of incorrect selection of mismatches caused by GC-rich tags is avoided.

Dwg.0/3

L116 ANSWER 22 OF 22 WPIDS (C) 2003 THOMSON DERWENT

ACCESSION NUMBER: 1996-222001 [22] WPIDS

CROSS REFERENCE: 1996-222023 [22]; 1997-099943 [09]; 1997-235911 [21]; 1998-042210 [04]; 1999-024716 [02]; 2000-170257 [14]

DOC. NO. CPI: C1996-070480

TITLE: Repertoire of oligo nucleotide tags comprise molecular tagging system - used to track, identify and/or sort mols., e.g. polynucleotide(s), for large scale **DNA** sequencing, etc..

DERWENT CLASS: B04 D16

INVENTOR(S): **BRENNER, S**; MACEVICZ, S C

PATENT ASSIGNEE(S): (LYNX-N) LYNX THERAPEUTICS INC; (SPEC-N) SPECTRAGEN INC

COUNTRY COUNT: 27

PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 9612014	A1	19960425	(199622)*	EN	61
RW: AT BE CH DE DK ES FR GB GR IE IT LU MC NL PT SE					
W: AU CA CZ FI HU JP KR NO SG					
AU 9642778	A	19960506	(199636)		
US 5604097	A	19970218	(199713)		26
US 5635400	A	19970603	(199728)		24
NO 9701644	A	19970602	(199732)		
FI 9701473	A	19970604	(199736)		
US 5654413	A	19970805	(199737)		26
EP 793718	A1	19970910	(199741)	EN	
R: AT BE CH DE DK ES FR GB GR IE IT LI LU MC NL PT SE					
CZ 9700866	A3	19970917	(199743)		
JP 10507357	W	19980721	(199839)		70
KR 97707279	A	19971201	(199847)		
HU 77916	T	19981028	(199850)		
AU 712929	B	19991118	(200007)		
AU 9952663	A	19991209	(200009)		
US 6280935	B1	20010828	(200151)		

APPLICATION DETAILS:

PATENT NO	KIND		APPLICATION	DATE
WO 9612014	A1		WO 1995-US12791	19951012
AU 9642778	A		AU 1996-42778	19951012
US 5604097	A	CIP of	US 1994-322348	19941013
			US 1994-358810	19941219
US 5635400	A	CIP of	US 1994-322348	19941013
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			US 1995-478238	19950607
NO 9701644	A		WO 1995-US12791	19951012
			NO 1997-1644	19970410
FI 9701473	A		WO 1995-US12791	19951012
			FI 1997-1473	19970409
US 5654413	A	CIP of	US 1994-322348	19941013
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			US 1995-484712	19950607
EP 793718	A1		EP 1995-941325	19951012
			WO 1995-US12791	19951012
CZ 9700866	A3		WO 1995-US12791	19951012
			CZ 1997-866	19951012
JP 10507357	W		WO 1995-US12791	19951012
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KR 97707279	A		WO 1995-US12791	19951012
			KR 1997-702433	19970414
HU 77916	T		WO 1995-US12791	19951012
			HU 1998-1187	19951012
AU 712929	B		AU 1996-42778	19951012
AU 9952663	A	Div ex	AU 1996-42778	19951012
			AU 1999-52663	19991004
US 6280935	B1	CIP of	US 1994-322348	19941013
		CIP of	US 1994-358810	19941219
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		Div ex	US 1996-659453	19960606
			US 1998-90809	19980604

FILING DETAILS:

PATENT NO	KIND		PATENT NO
AU 9642778	A	Based on	WO 9612014
EP 793718	A1	Based on	WO 9612014
CZ 9700866	A3	Based on	WO 9612014
JP 10507357	W	Based on	WO 9612014
KR 97707279	A	Based on	WO 9612014
HU 77916	T	Based on	WO 9612014
AU 712929	B	Previous Publ.	AU 9642778
		Based on	WO 9612014
AU 9952663	A	Div ex	AU 712929
US 6280935	B1	CIP of	US 5604097
		Div ex	US 5846719

PRIORITY APPLN. INFO: US 1994-358810 19941219; US 1994-322348 19941013; US 1995-478238 19950607; US 1995-484712 19950607

AB WO 9612014 A UPAB: 20011001
Repertoire of oligonucleotide tags (molecular tagging system) of formula (I), are claimed, S1S2S3....Sn (I), S1-Sn is a subunit comprising an oligonucleotide of 3-6 bases from a minimally cross-hybridising set, n = 4-10.

USE - The repertoire of oligonucleotide tags of the formula (I) can be used to sort a mol. or subpopulation of mols. from a population of mols, determine the nucleotide sequence of a target polynucleotide, classify a population of polynucleotides, sort a mix. of polynucleotides and detect novel cDNA mols. in a cDNA **library** (all claimed). It can also be used to sort cloned and identically tagged **DNA** fragments onto distinct solid phase supports to allow direct sequencing of multiple samples in parallel. Other uses include construction of combinatorial chemical **libraries**, large-scale mapping of **DNA**, mRNA fingerprinting and medical diagnostics.
Dwg.0/5

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FILE COVERS 1907 - 5 Mar 2003  VOL 138 ISS 10
FILE LAST UPDATED: 4 Mar 2003  (20030304/ED)

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previously printed

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    GUIDES, PLEASE VISIT:
    http://www.derwent.com/userguides/dwpi_guide.html <<<

=> d que l68; d que l72

L57        13227 SEA FILE=WPIDS ABB=ON  LIBRAR?
L58       123232 SEA FILE=WPIDS ABB=ON  HETERO?
L59        10822 SEA FILE=WPIDS ABB=ON  DUPLEX?
L60          207 SEA FILE=WPIDS ABB=ON  HETERODUPLEX?
L61        68152 SEA FILE=WPIDS ABB=ON  DNA OR ?NUCLEIC ACID#
L62         1610 SEA FILE=WPIDS ABB=ON  ENDONUCLEASE# OR ENDO NUCLEASE#
L64         3452 SEA FILE=WPIDS ABB=ON  SINGLESTRAND? OR SINGLE STRAND?
L68            4 SEA FILE=WPIDS ABB=ON  L57 AND ((L58 AND L59) OR L60) AND L61
                AND L62 AND L64



L57        13227 SEA FILE=WPIDS ABB=ON  LIBRAR?
L58       123232 SEA FILE=WPIDS ABB=ON  HETERO?
L59        10822 SEA FILE=WPIDS ABB=ON  DUPLEX?
L60          207 SEA FILE=WPIDS ABB=ON  HETERODUPLEX?
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L72            3 SEA FILE=WPIDS ABB=ON  L57 AND (L58 OR L59 OR L60) AND L61 AND
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=> s (l68 or l72) not l65

L119          4 (L68 OR L72) NOT L65
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previously printed

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=> fil biotechds

FILE 'BIOTECHDS' ENTERED AT 10:18:53 ON 05 MAR 2003
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FILE LAST UPDATED: 24 FEB 2003       <20030224/UP>

>>> USE OF THIS FILE IS LIMITED TO BIOTECH SUBSCRIBERS <<<
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>>> NEW CLASSIFICATION SYSTEM FROM 2002 ONWARDS - SEE HELP CLA <<<

=> d que l93; s l93 not l83

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L76          1136 SEA FILE=BIOTECHDS ABB=ON  DUPLEX?
L77           343 SEA FILE=BIOTECHDS ABB=ON  HETERODUPLEX?
L78         96580 SEA FILE=BIOTECHDS ABB=ON  DNA OR (NUCLEIC OR DEOXYRIBONUCLEIC)
                  (W)ACID#
L79          5000 SEA FILE=BIOTECHDS ABB=ON  ENDONUCLEASE# OR ENDO NUCLEASE#
L80           212 SEA FILE=BIOTECHDS ABB=ON  (EXO OR EXONUCLEASE#)(1W)(III OR 3)

L81          3171 SEA FILE=BIOTECHDS ABB=ON  SINGLESTRAND? OR SINGLE STRAND?
L85             7 SEA FILE=BIOTECHDS ABB=ON  L74 AND (L75 OR L76 OR L77) AND L78
                  AND L79 AND L81
L87             4 SEA FILE=BIOTECHDS ABB=ON  L74 AND L78 AND L80 AND (L75 OR L76
                  OR L77)
L88             5 SEA FILE=BIOTECHDS ABB=ON  L74 AND L78 AND L80 AND L79
L91            13 SEA FILE=BIOTECHDS ABB=ON  L87 OR L88 OR L85
L92         38881 SEA FILE=BIOTECHDS ABB=ON  (DNA OR LIBRAR?)/TI
L93             9 SEA FILE=BIOTECHDS ABB=ON  L91 AND L92


L120            8 L93 NOT L83 previously printed

=> fil biosis

FILE 'BIOSIS' ENTERED AT 10:19:02 ON 05 MAR 2003
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FILE COVERS 1969 TO DATE.
CAS REGISTRY NUMBERS AND CHEMICAL NAMES (CNs) PRESENT
FROM JANUARY 1969 TO DATE.

RECORDS LAST ADDED: 26 February 2003 (20030226/ED)


=> d que l105; d que l107; d que l108; d que l109; d que l112

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L96        717927 SEA FILE=BIOSIS ABB=ON  DNA OR ?NUCLEIC ACID#
L97        312200 SEA FILE=BIOSIS ABB=ON  HETERO?
L98         15293 SEA FILE=BIOSIS ABB=ON  DUPLEX?
L99          3106 SEA FILE=BIOSIS ABB=ON  HETERODUPLEX?
L102        28579 SEA FILE=BIOSIS ABB=ON  SINGLESTRAND? OR SINGLE STRAND?
L105            4 SEA FILE=BIOSIS ABB=ON  L95 AND L96 AND L102 AND ((L97 AND
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L96        717927 SEA FILE=BIOSIS ABB=ON  DNA OR ?NUCLEIC ACID#
L101         1032 SEA FILE=BIOSIS ABB=ON  (EXO OR EXONUCLEASE#)(1W)(III OR 3)
L102        28579 SEA FILE=BIOSIS ABB=ON  SINGLESTRAND? OR SINGLE STRAND?
L107            2 SEA FILE=BIOSIS ABB=ON  L95 AND L96 AND L102 AND L101


L95         45266 SEA FILE=BIOSIS ABB=ON  LIBRAR?
L96        717927 SEA FILE=BIOSIS ABB=ON  DNA OR ?NUCLEIC ACID#
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L98       15293 SEA FILE=BIOSIS ABB=ON  DUPLEX?
L99        3106 SEA FILE=BIOSIS ABB=ON  HETERODUPLEX?
L101       1032 SEA FILE=BIOSIS ABB=ON  (EXO OR EXONUCLEASE#)(1W)(III OR 3)
L108          1 SEA FILE=BIOSIS ABB=ON  L95 AND L96 AND (L97 OR L98 OR L99)
                AND L101


L95       45266 SEA FILE=BIOSIS ABB=ON  LIBRAR?
L96      717927 SEA FILE=BIOSIS ABB=ON  DNA OR ?NUCLEIC ACID#
L97      312200 SEA FILE=BIOSIS ABB=ON  HETERO?
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L99        3106 SEA FILE=BIOSIS ABB=ON  HETERODUPLEX?
L100      24889 SEA FILE=BIOSIS ABB=ON  ENDONUCLEASE# OR ENDO NUCLEASE#
L102      28579 SEA FILE=BIOSIS ABB=ON  SINGLESTRAND? OR SINGLE STRAND?
L109          2 SEA FILE=BIOSIS ABB=ON  L95 AND L96 AND L102 AND (L97 OR L98
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L95       45266 SEA FILE=BIOSIS ABB=ON  LIBRAR?
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L97      312200 SEA FILE=BIOSIS ABB=ON  HETERO?
L98       15293 SEA FILE=BIOSIS ABB=ON  DUPLEX?
L99        3106 SEA FILE=BIOSIS ABB=ON  HETERODUPLEX?
L100      24889 SEA FILE=BIOSIS ABB=ON  ENDONUCLEASE# OR ENDO NUCLEASE#
L101       1032 SEA FILE=BIOSIS ABB=ON  (EXO OR EXONUCLEASE#)(1W)(III OR 3)
L102      28579 SEA FILE=BIOSIS ABB=ON  SINGLESTRAND? OR SINGLE STRAND?
L112          1 SEA FILE=BIOSIS ABB=ON  L95 AND L96 AND L100 AND L101 AND
                ((L97 OR L98 OR L99) OR L102)

=> s (l105 or l107 or l108 or l109 or l112) not l103

L121          9 (L105 OR L107 OR L108 OR L109 OR L112) NOT L103
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previously printed

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FILE 'BIOTECHNO' ENTERED AT 10:19:29 ON 05 MAR 2003
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FILE 'BIOTECHDS' ENTERED AT 10:19:29 ON 05 MAR 2003
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FILE 'CAPLUS' ENTERED AT 10:19:29 ON 05 MAR 2003
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FILE 'BIOSIS' ENTERED AT 10:19:29 ON 05 MAR 2003
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FILE 'WPIDS' ENTERED AT 10:19:29 ON 05 MAR 2003
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PROCESSING COMPLETED FOR L118
PROCESSING COMPLETED FOR L120
PROCESSING COMPLETED FOR L117
PROCESSING COMPLETED FOR L121
PROCESSING COMPLETED FOR L119
L122         42 DUP REM L118 L120 L117 L121 L119 (6 DUPLICATES REMOVED)
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                ANSWERS '14-21' FROM FILE BIOTECHDS
                ANSWERS '22-34' FROM FILE CAPLUS
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ANSWERS '35-38' FROM FILE BIOSIS
ANSWERS '39-42' FROM FILE WPIDS

=> d ibib ab hitrn 1-42; fil hom

L122 ANSWER 1 OF 42 BIOTECHNO COPYRIGHT 2003 Elsevier Science B.V.DUPLICATE
ACCESSION NUMBER: 2000:30662585 BIOTECHNO
TITLE: Analysis of variability of P1 gene region of N strain of potato virus Y using temperature-gradient gel electrophoresis and **DNA heteroduplex** analysis
AUTHOR: Matousek J.; Ptacek J.; Dedic P.; Schubert J.
CORPORATE SOURCE: J. Matousek, Department of Molecular Genetics, Institute of Plant Molecular Biology, Czech Academy of Sciences, Branisovska 31, 370 05 Ceske Budejovice, Czech Republic.
E-mail: jmat@umbr.cas.cz
SOURCE: Acta Virologica, (2000), 44/1 (40-46), 32 reference(s)
CODEN: AVIRA2 ISSN: 0001-723X
DOCUMENT TYPE: Journal; Article
COUNTRY: Slovakia
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Reaction conditions specific for reverse taranscription-polymerase chain reaction (RT-PCR) of potato virus Y strain NTN (PVY(NTN)) were used to amplify a 394 bp fragment of the P1 gene from selected PVY isolates with the aim to study the PVY variability within this genomic region. The P1 gene fragment from the Nicola isolate (Nicola P1/1 clone) was sequenced and characterized by temperature-gradient gel electrophoresis (TGGE). The Nicola P1/1 clone differed from that from the Hungarian isolate by double point mutation resulting in two changes at the deduced amino acid level. The clone showed simple transition from double-stranded to **single -stranded** form with two characteristic melting end points of about 41.degree.C and 48.degree.C. A more complicated TGGE pattern was, however, found for the whole P1 cDNA **library** of the Nicola isolate, suggesting accumulation of some minor sequence variants of PVY in this isolate. Based on the TGGE pattern, 46.degree.C was selected as the standard temperature for electrophoretic analysis of **heteroduplex** DNAs formed with the Nicola P1/1 **DNA** as reference. More than 40 other PVY isolates from PVY(N) group were analysed using this method. In most cases only minor fractions of electrophoretically distinguishable **DNA heteroduplexes** were found, however, in most isolates of PVY(N)-Wilga type, mixtures of the major sequence variants were observed. Two of these variants from the hybrid 220-5 (Czech Republic) were sequenced. Both of them differed from the Nicola P1/1 clone by 6 point mutations, which led to several changes at the amino acid level.

L122 ANSWER 2 OF 42 BIOTECHNO COPYRIGHT 2003 Elsevier Science B.V.DUPLICATE
ACCESSION NUMBER: 1999:29393237 BIOTECHNO
TITLE: Identification and characterization of the molecular lesion causing mucopolysaccharidosis type I in cats
AUTHOR: He X.; Li C.-M.; Simonaro C.M.; Wan Q.; Haskins M.E.; Desnick R.J.; Schuchman E.H.
CORPORATE SOURCE: E.H. Schuchman, Department of Human Genetics, Box 1498, Mount Sinai School of Medicine, 100th Street and Fifth Avenue, New York, NY 10029, United States.
E-mail: Schuchman@msvax.mssm.edu
SOURCE: Molecular Genetics and Metabolism, (1999), 67/2 (106-112), 26 reference(s)
CODEN: MGMEFF ISSN: 1096-7192
DOCUMENT TYPE: Journal; Article
COUNTRY: United States

LANGUAGE: English
SUMMARY LANGUAGE: English

AB Mucopolysaccharidosis Type I (MPS I) is the lysosomal storage disease caused by the deficient activity of .alpha.-L-iduronidase (IDUA). In man, MPS I can occur in severe, mild, or intermediate forms known as the Hurler, Scheie, or Hurler/Scheie syndromes, respectively. MPS I also has been described in cats, dogs, and mice. This manuscript reports the identification and characterization of the mutation causing MPS I in cats. To obtain wild-type feline IDUA cDNAs, two PCR-based strategies were used. PCR primers were constructed from a conserved region of the published human and dog sequences and used to amplify a 224-bp IDUA fragment from normal cat genomic **DNA**. This fragment was then used to screen a feline uterus cDNA **library**. PCR also was used to directly amplify IDUA fragments from the same cDNA **library**. Two overlapping feline IDUA cDNAs encoding 466 amino acid residues of the feline IDUA polypeptide (.sim.85% of the mature protein based on comparison to the human, dog, and mouse sequences) were obtained by these strategies. To identify the mutation causing MPS I in cats, **DNA** sequencing was carried out on the corresponding IDUA region from several affected animals. A 3-bp deletion was found on both IDUA alleles in each of the MPS I animals, predicting the deletion of a single aspartate residue from the feline IDUA polypeptide. To confirm the authenticity of this mutation, **heteroduplex**, SSCP, and transient expression studies were carried out. Over 100 animals from the MPS I colony were screened for the presence of the mutation by **heteroduplex** and SSCP analyses - in all cases the presence of the 3-bp deletion was 100% concordant with the disease phenotype. For transient expression studies, the two partial, overlapping feline cDNAs were combined and joined in-frame to the 5' end of the canine IDUA cDNA. This wild-type, hybrid cDNA expressed IDUA activity up to sixfold over endogenous levels after transfection into COS-1 cells. A modified full-length IDUA cDNA containing the 3-bp deletion did not express IDUA activity in a transient expression system, providing proof that this lesion was the cause of feline MPS I.

L122 ANSWER 3 OF 42 BIOTECHNO COPYRIGHT 2003 Elsevier Science B.V.DUPLICATE
ACCESSION NUMBER: 1992:22231582 BIOTECHNO
TITLE: cDNA cloning, sequencing, expression and possible domain structure of human APEX nuclease homologous to Escherichia coli **exonuclease III**
AUTHOR: Seki S.; Hatsushika M.; Watanabe S.; Akiyama K.; Nagao K.; Tsutsui K.
CORPORATE SOURCE: Department of Molecular Biology, Inst. of Cellular/Molecular Biology, Okayama University Medical School, 2-5-1, Shikata-cho,Okayama 700, Japan.
SOURCE: Biochimica et Biophysica Acta - Gene Structure and Expression, (1992), 1131/3 (287-299)
CODEN: BBGSD5 ISSN: 0167-4781
DOCUMENT TYPE: Journal; Article
COUNTRY: Netherlands
LANGUAGE: English
SUMMARY LANGUAGE: English

AB cDNA encoding the human homologue of mouse APEX nuclease was isolated from a human bone-marrow cDNA **library** by screening with cDNA for mouse APEX nuclease. The mouse enzyme has been shown to possess four enzymatic activities, i.e., apurinic/apyrimidinic endonuclease, 3'-5' exonuclease, **DNA** 3'-phosphatase and **DNA** 3' repair diesterase activities. The cDNA for human APEX nuclease was 1420 nucleotides long, consisting of a 5' terminal untranslated region of 205 nucleotide long, a coding region of 954 nucleotide long encoding 318 amino acid residues, a 3' terminal untranslated region of 261 nucleotide long, and a poly(A) tail. Determination of the N-terminal amino acid sequence of APEX nuclease purified from HeLa cells showed that the mature

enzyme lacks the N-terminal methionine. The amino acid sequence of human APEX nuclease has 94% sequence identity with that of mouse APEX nuclease, and shows significant homologies to those of Escherichia coli **exonuclease III** and Streptococcus pneumoniae ExoA protein. The coding sequence of human APEX nuclease was cloned into the pUC18 SmaI site in the control frame of the lacZ promoter. The construct was introduced into BW2001 (xth-11, nfo-2) strain and BW9109 (.DELTA.xth) strain cells of E. coli. The transformed cells expressed a 36.4 kDa polypeptide (the 317 amino acid sequence of APEX nuclease headed by the N-terminal decapeptide derived from the part of pUC18 sequence), and were less sensitive to methylmethanesulfonate and tert-butyl-hydroperoxide than the parent cells. The N-terminal regions of the constructed protein and APEX nuclease were cleaved frequently during the extraction and purification processes of protein to produce the 31, 33 and 35 kDa C-terminal fragments showing priming activities for **DNA** polymerase on acid-depurinated **DNA** and bleomycin-damaged **DNA**. Formation of such enzymatically active fragments of APEX nuclease may be a cause of **heterogeneity** of purified preparations of mammalian AP endonucleases. Based on analyses of the deduced amino acid sequence and the active fragments of APEX nuclease, it is suggested that the enzyme is organized into two domains, a 6 kDa N-terminal domain having nuclear location signals and 29 kDa C-terminal, catalytic domain.

L122 ANSWER 4 OF 42 BIOTECHNO COPYRIGHT 2003 Elsevier Science B.V.DUPLICATE

ACCESSION NUMBER:	1988:24315868 BIOTECHNO
TITLE:	cDNA cloning of the immunoglobulin heavy chain binding protein
AUTHOR:	Haas I.G.; Meo T.
CORPORATE SOURCE:	Unite d'Immunogenetique, Institut Pasteur,75724 Paris Cedex 15, France.
SOURCE:	Proceedings of the National Academy of Sciences of the United States of America, (1988), 85/7 (2250-2254) CODEN: PNASA6 ISSN: 0027-8424
DOCUMENT TYPE:	Journal; Article
COUNTRY:	United States
LANGUAGE:	English
SUMMARY LANGUAGE:	English

AB A cDNA **library** was constructed from size-fractionated poly(A).sup.+ RNA prepared from a murine pre-B-cell bybridoma expressing high levels of immunoglobulin heavy chain binding protein (BiP) and .mu. heavy chains. Transformed bacterial colonies were screened for recombinant plasmids containing cDNA coding for BiP by hybrid-selected mRNA translation. A clone, pMBiP, containing a 736-base-pair insert was shown to encode the protein. Translation in vitro of hybridoma mRNA selected by hybridization to the pMBiP cDNA yielded a single polypeptide of BiP-like size. The authenticity of this mRNA was verified by comparing the peptides obtained by the limited proteolysis of its in vitro translation product with those obtained from the in vivo produced BiP. Likewise, the authenticity of the cDNA insert was verified by an RNase A protection assay of **heteroduplex** molecules obtained by annealing a uniformly labeled **single-strand** copy of the cDNA clone with the same mRNA selected by hybridization and tested by translation. The nucleotide sequence of this clone enabled us to deduce the carboxyl-terminal 142 amino acids of BiP and to establish its kinship with the 70-kDa heat shock protein family. The finding of a single copy of the BiP gene in **DNA** blots of mouse and rat implies that the BiP-related RNA transcripts constitutively expressed in various murine tissues and cell lines are indeed products of the same gene. These findings imply that BiP plays a more general role than previously anticipated on the basis of the discovery of its association with immunoglobulin heavy chains.

L122 ANSWER 5 OF 42 BIOTECHNO DUPLICATE
ACCESSION NUMBER: 1983:14244248 BIOTECHNO
TITLE: Physical characterization and molecular cloning of the Shope fibroma virus **DNA** genome
AUTHOR: Wills A.; Delange A.M.; Gregson C.; et al.
CORPORATE SOURCE: Department of Biochemistry, University of Alberta, Edmonton, Alta., Canada.
SOURCE: Virology, (1983), 130/2 (403-414)
CODEN: VIRLAX
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English

AB **DNA** from several independent strains of Shope fibroma virus, a tumorogenic leporipoxvirus of rabbits, was isolated and analyzed by restriction **endonuclease** digestion and Southern blotting. The restriction profiles indicated a high degree of sequence conservation among the isolates but blotting under standard stringencies revealed no detectable cross homology with a member of the orthopoxvirus group, vaccinia. The genome of the fibroma virus was calculated to be in excess of 160 kilobases and shown to possess two features analogous to the orthopoxvirus group: (1) the terminal restriction fragments possess covalently closed hairpin structures; and (2) the terminal sequences are present as inverted repeats of greater than 10 kilobases. The terminal 3.6 kilobase BamHI restriction fragment was cloned in pBR322 after removal of the hairpin structure with mung bean **single strand**-specific **endonuclease** and addition of BamHI linkers. SFV sequences within this terminal region were shown, using .sup.3.sup.2P SFV cloned terminal probe, to have none of the sequence **heterogeneity** characteristic of vaccinia **DNA** termini.
The remaining 20 internal SFV BamHi restriction fragments were propagated in bacterial plasmids either as intact fragments, or after secondary digestion with HindIII, and together constitute the complete cloned SFV sequence **library**.

L122 ANSWER 6 OF 42 BIOTECHNO
ACCESSION NUMBER: 2002:34968914 BIOTECHNO
TITLE: NM23-H1 and NM23-H2 repress transcriptional activities of **nuclease**-hypersensitive elements in the platelet-derived growth factor-A promoter
AUTHOR: Ma D.; Xing Z.; Liu B.; Pedigo N.G.; Zimmer S.G.; Bai Z.; Postel E.H.; Kaetzel D.M.
CORPORATE SOURCE: D.M. Kaetzel, Dept. of Biomedical Pharmacology, Univ. of Kentucky Medical Center, MS305, 800 Rose St., Lexington, KY 40536, United States.
E-mail: dmkaetz@pop.uky.edu
SOURCE: Journal of Biological Chemistry, (11 JAN 2002), 277/2 (1560-1567), 59 reference(s)
CODEN: JBCHA3 ISSN: 0021-9258
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
SUMMARY LANGUAGE: English

AB The platelet-derived growth factor (PDGF)-A promoter is regulated by a number of GC-rich regulatory elements that possess non-B-form **DNA** structures. Screening of a HeLa cDNA expression **library** with the C-rich strand of a PDGF-A silencer sequence (5'-S1 **nuclease** -hypersensitive site (SHS)) yielded three cDNA clones encoding NM23-H1, a protein implicated as a suppressor of metastasis in melanoma and breast carcinoma. Recombinant human NM23-H1 cleaved within the 3'-portions of both 5'-SHS strands in either **single-stranded** or **duplex** forms. In contrast, NM23-H2, known as a transcriptional activator with a **DNA** cleavage function, cleaved within the 5'-portions of both strands, revealing that NM23-H1 and NM23-H2 cleave at

distinct sites of the 5'-SHS and by different mechanisms. NM23-H1 and NM23-H2 also cleaved within the PDGF-A basal promoter region, again exhibiting preferences for cleavage within the 5'- and 3'-portions of the element, respectively. Transient transfection analyses in HepG2 cells revealed that both NM23-H1 and -H2 repressed transcriptional activity driven by the PDGF-A basal promoter (-82 to +8). Activity of the negative regulatory region (-1853 to -883), which contains the 5'-SHS, was also inhibited modestly by NM23-H1 and NM23-H2. These studies demonstrate for the first time that NM23-H1 interacts both structurally and functionally with **DNA**. They also indicate a role for NM23 proteins in repressing transcription of a growth factor oncogene, providing a possible molecular mechanism to explain their metastasis-suppressing effects.

L122 ANSWER 7 OF 42 BIOTECHNO COPYRIGHT 2003 Elsevier Science B.V.
ACCESSION NUMBER: 1999:29486572 BIOTECHNO
TITLE: Incremental truncation as a strategy in the engineering of novel biocatalysts
AUTHOR: Ostermeier M.; Nixon A.E.; Benkovic S.J.
CORPORATE SOURCE: S.J. Benkovic, Department of Chemistry, Pennsylvania State University, University Park, PA 16802, United States.
E-mail: sjb1@psu.edu
SOURCE: Bioorganic and Medicinal Chemistry, (1999), 7/10 (2139-2144), 19 reference(s)
CODEN: BMECEP ISSN: 0968-0896
PUBLISHER ITEM IDENT.: S0968089699001431
DOCUMENT TYPE: Journal; General Review
COUNTRY: United Kingdom
LANGUAGE: English
SUMMARY LANGUAGE: English
AB The application and success of combinatorial approaches to protein engineering problems have increased dramatically. However, current directed evolution strategies lack a combinatorial methodology for creating libraries of hybrid enzymes which lack high homology or for creating libraries of highly homologous genes with fusions at regions of non-identity. To create such hybrid enzyme libraries, we have developed a series of combinatorial approaches that utilize the incremental truncation of genes, gene fragments or gene libraries. For incremental truncation, **Exonuclease III** is used to create a library of all possible single base-pair deletions of a given piece of **DNA**. Incremental truncation libraries (ITLs) have applications in protein engineering as well as protein folding, enzyme evolution, and the chemical synthesis of proteins. In addition, we are developing a methodology of **DNA** shuffling which is independent of **DNA** sequence homology.

L122 ANSWER 8 OF 42 BIOTECHNO COPYRIGHT 2003 Elsevier Science B.V.
ACCESSION NUMBER: 1999:29365108 BIOTECHNO
TITLE: Determining the influence of structure on hybridization using oligonucleotide arrays
AUTHOR: Mir K.U.; Southern E.M.
CORPORATE SOURCE: K.U. Mir, Department of Biochemistry, University of Oxford, South Parks Rd., Oxford OX1 3QU, United Kingdom.
E-mail: kalim@bioch.ox.ac.uk
SOURCE: Nature Biotechnology, (1999), 17/8 (788-792), 56 reference(s)
CODEN: NABIF0 ISSN: 1087-0156
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
SUMMARY LANGUAGE: English

AB We have studied the effects of structure on **nucleic acid heteroduplex** formation by analyzing hybridization of tRNA(phe) to a complete set of complementary oligonucleotides, ranging from single nucleotides to dodecanucleotides. The analysis points to features in tRNA that determine **heteroduplex** yield. All **heteroduplexes** that give high yield include both double-stranded stems as well as **single-stranded** regions. Bases in the **single-stranded** regions are stacked onto the stems, and **heteroduplexes** terminate at potential interfaces for coaxial stacking. **Heteroduplex** formation is disfavored by sharp turns or a lack of helical order in **single- stranded** regions, competition from bases displaced from a stem, and stable tertiary interactions. The study is relevant to duplex formation on oligonucleotide microarrays and to antisense technologies.

L122 ANSWER 9 OF 42 BIOTECHNO

ACCESSION NUMBER: 1998:28412422 BIOTECHNO
TITLE: Design of a combinatorial oligonucleotide **library** containing all possible hexamer palindromes: PCR synthesis and application for identifying restriction cleavage sites
AUTHOR: Jadhav V.R.; Ganesh K.N.
CORPORATE SOURCE: K.N. Ganesh, Bioorganic Unit, Organic Chemistry Synthesis Division, National Chemical Laboratory, Pune 411008, India.
E-mail: kng@ems.ncl.res.in
SOURCE: Biochemical and Biophysical Research Communications, (14 JAN 1998), 242/2 (297-302), 20 reference(s)
CODEN: BBRCA0 ISSN: 0006-291X
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
SUMMARY LANGUAGE: English

AB An algorithm for designing a combinatorial **library** comprehensively representing all hexamer palindrome sequences at uniquely defined sites is described. The expected size for such a **library** of 64 possible hexamer palindromes is 384 bases, which is reduced to 266 bases spread over 8 oligonucleotides through a linear overlap of rationally selected hexamer palindromes. The **single stranded** oligonucleotides of the designed sets were chemically synthesized and converted into corresponding **duplex** dimers using PCR primer-dimer method. The utility of these **duplex** oligomers for identifying cleavage sites of restriction enzymes recognizing hexamer palindromes has been demonstrated using some representative enzymes. The **library** is also useful for screening restriction enzymes with tetramer cleavage sites and identifying the 'star' sites of restriction enzymes. The sets of oligonucleotides with high information content, though designed for direct and unambiguous characterization of cleavage sites of isolated restriction enzymes, have potential applications as templates for characterizing sequence selective binding and interaction of small molecules **nucleic acid**.

L122 ANSWER 10 OF 42 BIOTECHNO

ACCESSION NUMBER: 1995:25154453 BIOTECHNO
TITLE: An efficient strategy to isolate full-length cDNAs based on an mRNA cap retention procedure (CAPture)
AUTHOR: Edery I.; Chu L.L.; Sonenberg N.; Pelletier J.
CORPORATE SOURCE: Dept. of Biochemistry, McIntyre Medical Sciences Building, McGill University, 3655 Drummond St.,Montreal, Que. H3G 1Y6, Canada.
SOURCE: Molecular and Cellular Biology, (1995), 15/6 (3363-3371)

CODEN: MCEBD4 ISSN: 0270-7306
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
SUMMARY LANGUAGE: English
AB The ability to generate cDNA **libraries** is one of the most fundamental procedures in contemporary molecular biology. One of the major drawbacks of current methods is that most cDNAs present in any given **library** are incomplete, rendering the characterization of genes an inefficient and time- consuming task. We have developed an affinity selection procedure using a fusion protein containing the murine cap-binding protein (eukaryotic initiation factor 4E), coupled to a solid support matrix, that allows for the purification of mRNAs via the 5' cap structure. When combined with a **single- strand** -specific RNase digestion step, specific retention of complete cDNA- RNA **duplexes** following first-strand synthesis is achieved. This method can be used to generate cDNA **libraries** in which polyadenylated and nonpolyadenylated mRNAs are equally represented and to enrich for full-length or 5'-end clones, thus facilitating cDNA cloning and promoter mapping.

L122 ANSWER 11 OF 42 BIOTECHNO
ACCESSION NUMBER: 1994:24336408 BIOTECHNO
TITLE: Differential cDNA cloning by enzymatic degrading subtraction (EDS)
AUTHOR: Zeng J.; Gorski R.A.; Hamer D.
CORPORATE SOURCE: Laboratory of Biochemistry, National Cancer Institute, National Institutes of Health,Bethesda, MD 20892, United States.
SOURCE: Nucleic Acids Research, (1994), 22/21 (4381-4385)
CODEN: NARHAD ISSN: 0305-1048
DOCUMENT TYPE: Journal; Article
COUNTRY: United Kingdom
LANGUAGE: English
SUMMARY LANGUAGE: English
AB We describe a new method, called enzymatic degrading subtraction (EDS), for the construction of subtractive libraries from PCR amplified cDNA. The novel features of this method are that i) the tester **DNA** is blocked by thionucleotide incorporation; ii) the rate of hybridization is accelerated by phenol-emulsion reassociation; and iii) the driver cDNA and hybrid molecules are enzymatically removed by digestion with **exonucleases III** and VII rather than by physical partitioning. We demonstrate the utility of EDS by constructing a subtractive library enriched for cDNAs expressed in adult but not in embryonic rat brains.

L122 ANSWER 12 OF 42 BIOTECHNO
ACCESSION NUMBER: 1992:22240605 BIOTECHNO
TITLE: A new strategy useful for rapid identification of microsatellites from **DNA** libraries with large size inserts
AUTHOR: Baron B.; Poirier C.; Simon-Chazottes D.; Barnier C.; Guenet J.-L.
CORPORATE SOURCE: Unite de Genetique des Mammiferes de, Institut Pasteur, 25 rue du Docteur Roux,75724 Paris Cedex 15, France.
SOURCE: Nucleic Acids Research, (1992), 20/14 (3665-3669)
CODEN: NARHAD ISSN: 0305-1048
DOCUMENT TYPE: Journal; Article
COUNTRY: United Kingdom
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Microsatellites are new powerful polymorphic markers used for gene

mapping. Their characterization requires that all the sequence surrounding the repeat be known in order to be able to design primers for PCR amplification. However, when using **DNA** libraries with large cloned inserts, this sequence characterization is not immediately practicable. In this paper, we describe a new strategy, based both on the use of a microsatellite specific probing and on the creation of nested deleted clones with the **Exonuclease III**, in order to position microsatellites in a range allowing direct sequencing. This method was applied to the screening of a mouse chromosome 19 **DNA** specific library. In this way, thirteen clones were identified by specific probing and seven were submitted to the nested deletion strategy. Five of them presented microsatellite sequences in specific deleted subclones which were selected and sequenced. Primers were designed for each of them and polymorphism between the genomes of several inbred strain of mouse have been determined. These microsatellites were mapped, three of them to chromosome 19 and two to chromosome 11.

L122 ANSWER 13 OF 42 BIOTECHNO COPYRIGHT 2003 Elsevier Science B.V.
ACCESSION NUMBER: 1991:21315762 BIOTECHNO
TITLE: Isolation of cDNA clones encoding a human apurinic/apyrimidinic endonuclease that corrects **DNA** repair and mutagenesis defects in E.Coli xth (**exonuclease III**) mutants
AUTHOR: Robson C.N.; Hickson I.D.
CORPORATE SOURCE: Inst of Molecular Medicine, University of Oxford, John Radcliffe Hospital,Oxford OX3 9DU, United Kingdom.
SOURCE: Nucleic Acids Research, (1991), 19/20 (5519-5523)
CODEN: NARHAD ISSN: 0305-1048
DOCUMENT TYPE: Journal; Article
COUNTRY: United Kingdom
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Apurinic/apyrimidinic (AP) sites in cellular **DNA** are considered to be both cytotoxic and mutagenic, and can arise spontaneously or following exposure to **DNA** damaging agents. We have isolated cDNA clones which encode an endonuclease, designated HAP1 (human AP endonuclease 1), that catalyses the initial step in AP site repair in human cells. The predicted HAP1 protein has an M(r) of 35, 500 and shows striking sequence similarity (93% identity) to BAP 1, a bovine AP endonuclease enzyme. Significant sequence homology to two bacterial **DNA** repair enzymes, E. coli **exonuclease III** and S. pneumoniae ExoA proteins, and to Drosophila Rrp1 protein is also apparent. We have expressed the HAP1 cDNA in E. coli mutants lacking **exonuclease III** (xth), endonuclease IV (nfo), or both AP endonucleases. The HAP1 protein can substitute for **exonuclease III**, but not for endonuclease IV, in respect of some, but not all, **DNA** repair and mutagenesis functions. Moreover, a dut xth (ts) double mutant, which is nonviable at 42.degree.C due to an accumulation of unrepaired AP sites following excision of uracil from **DNA**, was rescued by expression of the HAP1 cDNA. These results indicate that AP endonucleases show remarkable conservation of both primary sequence and function. We would predict that the HAP1 protein is important in human cells for protection against the toxic and mutagenic effects of **DNA** damaging agents.

L122 ANSWER 14 OF 42 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI
ACCESSION NUMBER: 1999-02520 BIOTECHDS
TITLE: Method of normalizing cDNA **libraries**; cDNA **library** normalization and subtractive cDNA **library** construction using a directional cDNA **library**
AUTHOR: Bento Soares M; de Fatima Bonaldo M
PATENT ASSIGNEE: Univ.New-York-Columbia

LOCATION: New York, NY, USA.
PATENT INFO: US 5846721 8 Dec 1998
APPLICATION INFO: US 1996-715941 19 Sep 1996
PRIORITY INFO: US 1996-715941 19 Sep 1996
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 1999-059042 [05]
AB A method of normalizing a cDNA **library** is claimed and comprises: providing a cloned directional **library** (constructed in plasmid pT7T3-Pac vector) containing cDNA inserts in circular ds form capable of being amplified by polymerase chain reaction (generated using a **DNA** primer having a rare restriction **endonuclease** (**exonuclease III**) site for the first strand cDNA synthesis, especially a NotI or PacI site); converting the ds cDNA to ss **DNA** circles by transforming the cDNA **library** into Escherichia coli DH5alphaF and infecting it with helper phage M13K07 capable of carrying out the conversion; generating ss **nucleic acid** complementary to the circles with appropriate **DNA** primers and melting the ds cDNA inserts; hybridizing the ss **DNA** circles with the complementary ss **nucleic acid** to produce partial **duplexes**; and separating unhybridized ss **DNA** circles from hybridized circles by hydroxyapatite column chromatography. Also claimed are: a second method of normalizing a cDNA **library**; and a method to construct a subtractive cDNA **library**. (28pp)

L122 ANSWER 15 OF 42 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI
ACCESSION NUMBER: 2003-03145 BIOTECHDS
TITLE: Orientation-directed construction of a construct comprises performing polymerase chain reaction amplification using combined ligated sequences as a template and enrichment primers directing the amplification towards the desired orientation;
mutated **DNA** vector generation, polymerase chain reaction and restriction enzyme
AUTHOR: BEN-ASOULI Y; OSMAN F
PATENT ASSIGNEE: GENE BIO APPL LTD
PATENT INFO: WO 2002064774 22 Aug 2002
APPLICATION INFO: WO 2002-IL104 11 Feb 2002
PRIORITY INFO: IL 2001-141392 12 Feb 2001; IL 2001-141392 12 Feb 2001
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2002-667004 [71]
AB DERWENT ABSTRACT:
NOVELTY - Orientation-directed construction of a construct comprising at least 2 **nucleic acid** segments of interest, where each combined ligated sequence is amplified in a separate reaction creating a combined product having phosphorylated blunt ends, is new.
DETAILED DESCRIPTION - A method for orientation-directed construction of a construct comprising at least 2 **nucleic acid** segments of interest, comprising: (a) providing products having phosphorylated blunt ends derived from the **nucleic acid** segments of interest; (b) performing separate ligation reactions, where in each reaction the phosphorylated blunt-ended products of 2 different segments are ligated to create a combined ligated sequence; (c) performing polymerase chain reaction (PCR) amplification reaction using the combined ligated sequences a template and specific enrichment primers directing the PCR amplification towards the desired orientation, where each combined ligated sequence is amplified in a separate reaction creating a combined product having phosphorylated blunt ends; (d) isolating and purifying the combined product having the enriched desired orientation; (e) optionally, cyclic repetition of steps (b)-(d) for a desired number of times to create the isolated combined

product having the desired orientation; and (f) performing self-ligation of the isolated purified combined product obtained in (e) to create a circular double-stranded **DNA** construct containing the desired segments properly aligned and operably linked.

BIOTECHNOLOGY - Preferred Method: The first segment of interest comprises a replicable segment and the first segment of interest and/or any other segment of interest comprises a sequence coding for a selectable marker. The phosphorylated blunt ended products are obtained by: (i) performing a first set of PCR amplification using the segments as a template to create products having phosphorylated blunt ends, where each segment is amplified in a separate reaction using specific primers; (ii) creating blunt-ended products by cleaving the **nucleic acid** segments employing a blunt-end **endonuclease**; (iii) creating cohesive end products by cleaving the **nucleic acid** segments employing a sticky-end **endonuclease**, followed by fill-in reaction; and (iv) creating cohesive end products by cleavage of the **nucleic acid** segments employing a sticky-end **endonuclease**, followed by **single-strand DNA endonuclease** reaction and degrading **single-strand** extensions from the **DNA** ends. The phosphorylated blunt-ended products are obtained by phosphorylation of any one of the blunt-ended PCR products in (i) or the PCR primers by employing T4 kinase. The construct comprises more than 2 **nucleic acid** segments. The method further comprises cyclic repetition of steps (a)-(d) to create the isolated combined products having the desired orientation, and performing steps (b)-(f) to create a circular double-stranded **DNA** construct containing the desired segments properly aligned and operably linked. The replicable segment comprises an origin of replication (ori) sequence, and is derived from a replicable vector selected from retroviral vectors, phage vectors, plasmid vectors, expression vectors, self replicating vectors, phagemid vectors, and yeast artificial chromosomes (YAC) vectors. The specific primers employed in the first set of PCR amplification reaction are: a 5' primer comprising a sequence derived from the 5' end of a segment of interest (sense primer); and a 3' primer which is complementary to the sequence of the 3' end of the specific segment to be amplified (antisense primers). The specific primers employed in the amplification of the combined ligated sequences are: a 5' primer comprising a sequence derived from the 5' end of the first segment, which is upstream to the second ligated segment in the combined ligated product (sense primer); and a 3' primer which is complementary to the 3' end sequence of the second ligated (antisense primers), of the combined ligated sequence. Each of the primers comprises about 4-200 nucleotides, preferably 8-30 nucleotides. Ligation reaction is performed by employing **DNA** ligase, while PCR amplification is performed by employing a high fidelity **DNA** polymerase. The construct comprises: (a) one of the segments of interest comprising an origin of replication sequence and a sequence coding for a selectable marker; (b) at least one additional segment comprising a **heterologous** or homologous coding **nucleic acid** sequence of interest or mutations, fragments or derivatives; and (c) optionally, at least one additional segment comprising **nucleic acid** sequences coding for expression, control, promoting and/or regulatory elements. The **heterologous** or homologous coding sequence of interest encodes a protein selected from reporter proteins, enzymes, hormones, growth factors, cytokines, structural proteins, and industrially applicable proteins, or is itself a therapeutic product. The **heterologous** coding sequence encodes a reporter protein consisting of a green fluorescent protein, luciferase, secreted alkaline phosphatase or beta-galactosidase. The construct is an expression vehicle. The whole method may be performed manually or automatically.

USE - The method is useful for generating constructs or genetic vectors, specifically for orientation-directed construction of a mutated

DNA construct having at least one mutation.

ADVANTAGE - Compared with previous methods, the new method of producing genetic constructs is simpler, more efficient, rapid and inexpensive. The method provides genetic vectors aligned with their sequences in the proper orientation without the need for extensive scanning.

EXAMPLE - Twenty micrograms of primers were phosphorylated with 50 units of T4 polynucleotide kinase in the presence of 1 mM of ATP, and polymerase chain reaction (PCR) was performed using 1 microM of each primer per 20 microl, 200 microM dNTP, 10 ng template **DNA** and 1.5 units of Pfu **DNA** polymerase. PCR products were separated in agarose gel and the specific segment of interest was extracted by standard electroelution methods. Ligation reaction was performed in 20 microl of 1xT4 **DNA** ligase buffer containing 10 ng of each PCR product, 500 nM ATP and 400 units of T4 **DNA** ligase. E. coli strain JM109 was transformed with 10 microl of the self-ligation reaction and plated on LB+Amp, incubated overnight, and plasmids were isolated by standard mini-preparation protocol. **DNA** was analyzed with restriction enzymes, and the plasmid was purified by standard Large-scale plasmid protocol. The first segment was created by performing PCR using a human T-cell cDNA **library** as a template, phosphorylated primers 335 and 336, and the high fidelity thermal stable polymerase Pfu **DNA** polymerase that generated blunt ended product. The second segment was created by performing PCR using human T-cell cDNA **library** as a template, phosphorylated primers 826 and 827. PCR products were separated in 1.5% agarose gel and desired products were extracted using standard electroelution methods. A combined product comprising both segments was created by performing ligation reaction with the isolated PCR products. Ligation reaction product was amplified separately by 4 PCR using phosphorylated flanking primers to generate a third **DNA** segment. PCR products were separated in agarose gel and the specific segment was extracted by electro-elution. The combined product having the desired orientation was analyzed using PvuII restriction enzyme. Resulting products had all 4 possible orientations. (60 pages)

L122 ANSWER 16 OF 42 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI

ACCESSION NUMBER: 2003-01967 BIOTECHDS

TITLE: New nucleotide concatemers comprising a cassette of nucleotide sequence, useful for controlling and coordinating expression of large numbers of **heterologous** genes in a single host cell;
concatamer comprising a **DNA** cassette useful for vector-mediated gene transfer and expression in host cell

AUTHOR: GOLDSMITH N; SORENSEN A M P S; NIELSEN S V S; NAESBY M

PATENT ASSIGNEE: EVOLVA BIOTECH AS

PATENT INFO: WO 2002059296 1 Aug 2002

APPLICATION INFO: WO 2002-DK55 25 Jan 2002

PRIORITY INFO: US 2001-301022 27 Jun 2001; DK 2001-127 25 Jan 2001

DOCUMENT TYPE: Patent

LANGUAGE: English

OTHER SOURCE: WPI: 2002-619169 [66]

AB DERWENT ABSTRACT:

NOVELTY - A nucleotide concatemer comprising in the 5' to 3' direction a cassette comprising a fully defined nucleotide sequence, is new.

DETAILED DESCRIPTION - A nucleotide concatemer comprising in the 5' to 3' direction a cassette (where the first and second cassette are different) of nucleotide sequence of the general formula (I): (rs2-SP-PR-X-TR-SP-rs1)n (I) where: rs1 and rs2 = a restriction site; SP = a spacer of at least 2 nucleotide bases; PR = a promoter capable of functioning in a cell; X = an expressible nucleotide sequence; TR = terminator; n at least 2. INDEPENDENT CLAIMS are also included for the following: (1) a method for concatenation by concatenating at least 2

cassettes of nucleotide sequences each cassette comprising a first sticky end, a spacer sequence, a promoter, an expressible nucleotide sequence, a terminator, a spacer sequence, and a second sticky end; (2) a cell comprising at least one concatemer of individual oligonucleotide cassettes, where each concatemer comprises an oligonucleotide of the formula (I) in a 5' to 3' direction; (3) a method for producing a transgenic cell by inserting into a host cell a concatemer comprising a **heterologous** nucleotide sequence having at least 2 genes, each controlled by a different promoter; (4) a primary vector comprising a nucleotide sequence cassette of the general formula (II): (RS1-RS2-SP-PR-CS-TR-SP-RS2'-RS1') (II) where: RS1 and RS1' = restriction sites; RS2 and RS2' = restriction sties different from RS1 and RS1'; SP = a spacer sequence of at least 2 nucleotides; PR = a promoter; and TR = a terminator; and CS = cloning site; (5) a method of preparing vector by inserting an expressible nucleotide sequence into a cloning site in a primary vector comprising a cassette consisting of a nucleotide sequence of the general formula (II) in a 5' to 3' direction; (6) a nucleotide **library** comprising at least 2 primary vectors, each comprising a nucleotide sequence of the formula (III): (RS1-RS2-SP-PR-X-TR-SP-RS2'-RS1') (III) where: X = an expressible nucleotide sequence; RS1 and RS1' = restriction sites; RS2 and RS2' = restriction sties different from RS1 and RS1'; SP = a spacer sequence of at least 2 nucleotides; PR = a promoter; and TR = a terminator; and (7) a method of preparing a nucleotide **library** by obtaining expressible nucleotide sequences, cloning them into cloning sites of a mixture of primary vectors consisting of a cassette having a nucleotide sequence of the general formula (II).

BIOTECHNOLOGY - Preferred Concatemer: The nucleotide sequence comprises a **DNA** sequence consisting of cDNA or genomic **DNA**, and may be **single stranded**, partly **single stranded** or double stranded. The nucleotide sequences are from at least one or two expression state(s). The rs1-rs2 restriction sites of at least 2 cassettes or essentially all cassettes are recognized by the same restriction enzyme are preferably identical, where all the cassettes are different. At least one cassette, preferably all the cassettes, comprises an intron between the promoter and the expressible nucleotide sequence. The difference comprises different promoters, expressible nucleotide sequences, spacers, terminators, and/or introns. In (I), n is at least 10-2000. At least one cassette comprises the cassette from a primary vector. The concatemer is comprised in an artificial chromosome selected from a yeast, mega yeast, bacterial, mouse, mammalian, insect, avian, bacteriophage, baculovirus and human artificial chromosome. The concatemer may also be comprised in a plasmid or in an insertion vector, such as yeast integrative plasmid, yeast replicating plasmid, yeast episomal plasmid, yeast centromeric plasmid, yeast linear plasmid, yeast expression plasmid, yeast retrotransposons, yeast killer plasmid, or a yeast disintegration plasmid. The vector further comprises at least one selectable genetic marker, such as a repressive or a dominant marker, preferably 2 selectable genetic marker selected from LEU 2, TRP 1, HIS 3, LYS 2, URA 3, ADE 2, Amyloglucosidase, beta-lactamase, CUP 1, G418, TUN (RTM), KILk1, C230, SMR1, SFA, Hygromycin, methotrexate, chloramphenicol, Diuron, Zeocin (RTM), and Canavanine (RTM). The different expressible nucleotide sequences come from the same or different expression states, which represent at least 2 different tissues from at least 2 organs, species, or genera. The different species are from at least 2 different phyla, classes, divisions, sub-kingdoms, or kingdoms. The species is a eukaryote or prokaryote. The concatemer is designed to minimize the level of repeat sequences occurring in the concatemer. Preferred Method: Concatenation further comprises: (a) starting from a primary vector having a formula (III); (b) cutting the primary vector with at least one restriction enzyme specific for RS2 and RS2' to obtain cassettes having the general formula (IV), where rs1 and rs2 together denote a functional restriction

site RS2 or RS2; and (c) assembling the cut out cassettes through interaction between rs1 and rs2. The method comprises concatenating at least 10-2000 cassettes. Vector arms are added each having RS2 or RS2' in one end and a non-complementary overhang or a blunt end in the other end, where the ratio of vector arms to cassettes determines the number of cassettes in the concatemer, and the vector arms are preferably artificial chromosome vector arms. The method also includes adding stopper fragments each having RS2 or RS2' on one end and a non-complementary overhang or a blunt end in the other end, and ligating the vector arms to the stopper fragments. The mRNA is isolated from an expression state and full-length cDNA clones corresponding to the mRNA sequences are obtained. These full-length cDNA clones are inserted into a cloning site in a cassette in a primary vector, where the cassette is of the general formula (IV) (RS1-RS2-SP-PR-CS-TR-SP-RS2'-RS1') in 5' to 3' direction, where CS denotes a cloning site. The RS1 and RS1' are restriction sites leaving blunt ends, and are cleaved by one restriction enzyme. RS2 and RS2' are restriction sites leaving compatible sticky ends, and are cleaved by one restriction enzyme. RS1 and RS1' are identical, and RS2 and RS2' are also identical and have (non-)palindromic overhangs. The vector comprises selection vectors having expressible nucleotide sequences from at least two different expression states, such as from 2 different species, classes, divisions, sub-kingdoms or kingdoms. The concatemer is ligated into an artificial chromosome selected from yeast, mega yeast, bacterial, mouse and human artificial chromosome. Producing a transgenic cell further comprises selecting cells comprising at least stably maintained concatemer, by selecting cells carrying at least one selectable genetic marker on an artificial chromosome, preferably 2 selectable genetic markers on an artificial chromosome. The method of preparing a vector further comprises isolating total mRNA from an expression state, obtaining full-length cDNA for insertion into the vector, and selecting cDNA to obtain substantially full-length cDNA. Insertion into the primary vector is done by directional cloning. The substantially full-length cDNA comprises normalized representation of cDNA species characteristic of a given expression state. The nucleotide sequences are obtained from a cDNA **library**. Preferred Cell: The cell comprises 2-4 concatemers per cell. The eukaryotic cell is selected from yeasts, filamentous ascomycetes such as Neurospora crassa and Aspergillus nidulans; plant cells such as those derived from Nicotiana and Arabidopsis; mammalian host cells such as those derived from humans, monkeys and rodents; Chinese hamster ovary cells, NIH/3T3, COS, 293, Vero, and HeLa cells. The yeast cell can be further selected from baker's yeast, Kluyveromyces marxianus, K. lactis, Kluyveromyces, Candida utilis, C. paraffinica, C. shehatae, Candida spp. (C. palmioleophila), C. guilliermondii, C. brumptii, C. hydrocarbofumarica, C. tropicalis, C. flaveri, Phaffia rhodozyma, Saccharomyces boulardii, S. cerevisiae, Pichia pastoris, P. stipitis, Pichia spp., Hansenula polymorpha, Yarrowia lipolytica, Schwanniomyces castellii, Rhodotorula glutinis, Rhodotorula spp., Rodotorula rubra, Lypomyces lipofer, Cryptococcus curvatos, Saccharomycopsis spp., Aureobasidium pullulans, Torulopsis, Eremothecium ashbyii, Kloecker, Pachysolen spp. and Torulopsis bombicola. The cell has a mutation in a central biosynthetic pathway, and comprises an inserted selectable genetic marker complementing the mutation. The recombination within the expressible nucleotide sequence has been minimized. Preferred Vector: The nucleotide sequence of the vector is a double stranded **DNA**. The vector comprises an intron sequence between the promoter and the cloning site and/or between the cloning site and the terminator. The cloning site comprises an expressible nucleotide sequence which can be a full-length cDNA or a genomic **DNA**. The RS1, RS1', RS2, and RS2' comprise a rare restriction site, and has a recognition sequence comprising at least 6-10 bases. The recognition sequence comprises a bipartite sequence, and has a GC content of more than 40%, preferably more than 60%. The restriction enzyme recognizing RS2 and RS2' produces

sticky ends upon cleavage of a double stranded nucleotide sequence, where the sticky ends have pre-determined nucleotide sequence. The restriction enzyme recognizing RS1 and RS1' produces blunt ends upon cleavage of a double stranded nucleotide sequence, or produces sticky ends with a nucleotide sequence which is non-compatible with the nucleotide sequence of sticky ends produced upon cleavage of RS2 and RS2'. The vector comprises a spacer sequence between TR and RS2', where the spacer and the optional spacer sequence comprise at least 100-6000 bases, and the spacer sequences consists of 100-2500, preferably 700-1400 bases. The promoter is an externally controllable promoter, preferably selected from inducible or repressible promoter. The promoter is chemically inducible and/or repressible, and/or inducible and/or repressible by temperature. The promoter may also be induced and/or repressed by a factor selected from carbohydrates (e.g. galactose), low inorganic phosphate levels, temperature (e.g. low or high temperature shift), metals or metal ions (e.g. copper ions), hormones (e.g. dihydrotestosterone, or doexycorticosterone), heat shock (e.g. 39 degrees C), methanol, redox status, growth stage (e.g. developmental stage), and synthetic inducers (e.g. the gal inducer). The promoter is further selected from ADH1, PGK 1, GAP 491, TPI, PYK, ENO, PMA 1, PHO5, GAL 1, GAL2, GAL 10, MET25, ADH2, MEL 1, CUP 1, HSE, AOX, MOX, SV40, CaMV, Opaque-2, GRE, ARE, PGK/ARE hybrid, CYC/GRE hybrid, TIP/alpha2 operator, AOX 1, and MOX A. The promoter is preferably a synthetic promoter. The cloning site allows directional cloning, where the cloning site consists of multiple cloning sites, such as a polylinker site, and encodes a series of restriction **endonuclease** recognition sites. The promoter and terminator are capable of functioning in an expression host cell, preferably in a yeast cell. The primary vector comprising the cassette is a plasmid vector having a high copy number, capable of being propagated in E. coli, and having a selectable marker maintenance in E. coli. The primary vector can be made **single stranded**. The vector further includes an origin of replication in the vector backbone, preferably origin of replication for filamentous phages, specifically f1 origin of replication. The primary vector is selected from pBR322, pUC18, pUC19, pUC118, pUC119, pEMBL, pRSA101, and pBluescript. Preferred **Library**: The **library** is maintained in a host cell capable of maintaining the vectors having cassettes substantially unaltered. The host cell is a bacteria (e.g. E. coli or Bacillus subtillis), or fungi such as yeast. The promoters are not functional in the **library** host.

USE - The concatemer is useful for controlling and coordinating expression of large numbers of **heterologous** genes in a single host cell, and as powerful tools for generating novel and non-native combinations of genes for coordinated expression in a host cell. (124 pages)

L122 ANSWER 17 OF 42 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI

ACCESSION NUMBER: 2002-11778 BIOTECHDS
TITLE: Generating polynucleotide **libraries** having nucleotide deletions/additions at different positions in genetic element sequence by cleaving copies of circular polynucleotides and subjecting to removal or addition process ;
plasmid pLacZi-mediated beta-galactosidase gene transfer and expression in Escherichia coli
AUTHOR: SMIDER V
PATENT ASSIGNEE: INTEGRIGEN INC
PATENT INFO: WO 2002016642 28 Feb 2002
APPLICATION INFO: WO 2000-US25788 18 Aug 2000
PRIORITY INFO: US 2000-226477 18 Aug 2000
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2002-280947 [32]

AB DERWENT ABSTRACT:
NOVELTY - Multiple copies of circular polynucleotides (PN1) having a genetic element are subjected to random cleavage to obtain multiple linear PN1 each having at least one 3' and 5' end, and obtained linear PN1 is subjected to process which removes or adds one nucleotide from one of the ends to produce **library** of deletion/addition PN1 sequences with deletions/additions at different random positions.

DETAILED DESCRIPTION - Generating (M1) a **library** of PN1 having nucleotide deletions/additions at differing positions in a sequence of a genetic element by: (a) subjecting multiple copies of circular PN1 having the genetic element to random cleavage to obtain multiple linear PN1 each having at least one 3' and 5' end; and (b) subjecting linear PN1 to a process which removes at least one nucleotide from one of the end of PN1 producing a **library** of deletion/addition PN1 sequences with deletions/additions at different random positions. INDEPENDENT CLAIMS are also included for the following: (1) a substantially pure composition (I) comprising a **library** of multiple linear PN1 each having a different 3' and 5' end, where each linear PN1 is identical to the others if circularized; or comprising a **library** of at least two deletion/addition PN1 each differing from the other only by having a different random deletion/addition; (2) producing (M2) short deletions from the end of a polynucleotide by incubating a population of polynucleotides with an exonuclease at a temperature from 0-24 degreesC in the presence of 10-500 mM salt, thereby producing a population of polynucleotide containing deletions of 1-100 residues from at least one end of the polynucleotide; and (3) a substantially pure composition (II) of at least two polynucleotides each having two ends and differing from each other only by having different deletions of 1-100 residues at one or both ends; or of at least two polynucleotides each differing from one another only by deletions of 1-100 residues at a specific internal position within the polynucleotides.

BIOTECHNOLOGY - Preferred Method: M1 further comprises subjecting deletion/addition PN1 sequences to a process that covalently joins the 3' end and 5' end to one another and/or the **library** of PN1 thus obtained is further subjected to a process that selects for a desired function. Preferably the method further comprises repeating steps (a) and (b) and further includes a process for inserting nucleotides at the position of deletion or a process for deleting nucleotides at point of addition. In M1, the cleavage occurs with an **endonuclease** which is S1, and the **library** of deletion/addition PN1 comprises at least 5, preferably 10 and more preferably 30 individual PN1 each having a random deletion/addition at a different position from the others, where 1-3 or 50-100 nucleotides are deleted, or 1-3, 3-50 or more preferably 50-100 nucleotides are added. The composition of multiple copies of circular PN1 is free of naturally-occurring homologs to the genetic element. In M2, the polynucleotide is double exonuclease is **exonuclease III**, where the double-stranded **nucleic acid** is incubated with a single-stranded **endonuclease** which is S1 nuclease, to produce a blunt end. The resulting population of polynucleotides containing deletions at the ends are covalently joined to at least a second end producing a population of circular polynucleotides containing a deletion at an internal position, where the population of polynucleotides contains deletions of 1-50 residues from at least one end of the polynucleotide 1-50, preferably 1-30 residues from at least one end of the polynucleotide. Preferred Composition: (I) preferably comprises at least 5 polynucleotides having a different 3' and 5' end, each differing from the other only by having a different random deletion/addition, and deletion/addition PN1 further comprises at least one nucleotide inserted at the position of deletion, or addition. In (II), polynucleotides differ from one another by deletions of 1-50 preferably 1-30 and more preferably 1-10 residues at one or both ends or at the specific internal position.

USE - M1 is useful for generating a **library** of PN1 having nucleotide deletions/additions at differing positions in a sequence of a genetic element (claimed).

EXAMPLE - The plasmid pLacZi was propagated in DH10B Escherichia coli cells and plasmid was prepared by Qiagen maxiprep (RTM) columns. Plasmid **DNA** at 200 ng/mul was incubated with 0.4, 2.0, 10 or 50 units of S1 nuclease. The reaction was stopped and heated to 70 degreesC. Protein was removed by twice extracting with an equal volume of phenol:chloroform:isoamyl alcohol (I), once with an equal volume of ether, precipitated with sodium acetate and resuspended in water. Cleaved pLacZi was analyzed by 1.5% agarose gel electrophoresis. S1 nuclease cleaved plasmid was seen to co-migrate with pLacZi cleaved with ClaI, which cuts pLacZi once. Thus, S1 nuclease linearized a circular **DNA** molecule. To this end, linear plasmid produced by S1 cleavage was gel purified or purified and further cleaved with Cla I. Controls included supercoiled plasmid, plasmid linearized with Cla I or plasmid linearized with S1 nuclease and unpurified. CHO cell cDNA (5 mug) was fragmented with 0.001 units of DNase I. The reaction was stopped and heated. **DNA** was extracted with an equal volume of (I). Plasmid linearized with Cla I or S1 nuclease were dephosphorylated, then again extracted with an equal volume of phenol:chloroform:isoamyl alcohol, once with an equal volume of ether, and precipitated with sodium acetate. To insert random cDNA fragments into plasmid **DNA**, 0.2 mg of linearized, depehosphorylated plasmid was incubated with 1 ng of cDNA fragments in the presence of T4 **DNA** ligase (1.0 U). As controls, linearized plasmid was incubated with ligase in the absence of cDNA fragments, and cDNA fragments were incubated with ligase in the absence of linearized vector. DH10B Escherichia coli were then electroporated with 1.0 mul of each ligation mix. Several E.coli colonies were identified in the vector plus insert arms of the experiment which exhibited either white, intermediate, or blue phenotype on X-Gal plates. Polymerase Chain Reaction across the Cla I site in the colonies which arose from vector linearized with Cla I ligated to cDNA fragments revealed several clones containing inserts of sizes from 100-300 base pairs.(62 pages)

L122 ANSWER 18 OF 42 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI

ACCESSION NUMBER: 2002-11871 BIOTECHDS

TITLE: Subtractive hybridization for identifying sample polynucleotides, comprises blocking highly abundant sequences, removing low-abundance enriched sequences present in samples and producing test-specific **duplexes;**
sense or antisense **DNA** primer and **DNA** hybrid **duplex** formation for **DNA** detection, **DNA** synthesis and **DNA** array construction

AUTHOR: ZHU Y Y

PATENT ASSIGNEE: GENEMED BIOTECHNOLOGIES INC

PATENT INFO: WO 2002012564 14 Feb 2002

APPLICATION INFO: WO 2000-US24730 7 Aug 2000

PRIORITY INFO: US 2001-288777 4 May 2001

DOCUMENT TYPE: Patent

LANGUAGE: English

OTHER SOURCE: WPI: 2002-257486 [30]

AB DERWENT ABSTRACT:

NOVELTY - Subtractive hybridization (M1) for identifying polynucleotides in test samples that are absent, or less abundant, in reference sample comprising first subtractive hybridization to block high-abundance test sequences, leaving test strands enriched for low-abundance sequences, removing low-abundance enriched sequences present in both samples in second hybridization, and producing test-specific **duplexes**, is new.

DETAILED DESCRIPTION - Subtractive hybridization (M1) for

identifying polynucleotides in test samples that are absent, or less abundant, in reference sample comprising: (a) providing high-abundance enriched polynucleotide strands of, or prepared from, a pool of test or reference polynucleotides that is enriched in high-abundance polynucleotide sequences relative to a test or reference polynucleotide sample, respectively; (b) contacting the high-abundance enriched polynucleotide strands with test polynucleotide strands of, or prepared from, the test polynucleotide sample under hybridization conditions to form a first hybridization mixture, thus producing unhybridized test polynucleotide strands that are enriched in low-abundance polynucleotide sequences relative to the test polynucleotide sample; (c) synthesizing polynucleotide strands from the unhybridized test polynucleotide strands, thus producing low-abundance enriched test polynucleotide strands; (d) providing low-abundance-enriched reference polynucleotide strands of, or prepared from, a reference pool of polynucleotides that is enriched in low-abundance polynucleotide sequences relative to the reference polynucleotide sample; (e) contacting the low-abundance enriched reference polynucleotide strands with the low-abundance enriched test polynucleotide strands under hybridization conditions to form a second hybridization mixture, thus producing hybrid **duplexes**, unhybridized low-abundance enriched reference polynucleotide strands, and unhybridized low-abundance enriched test polynucleotide strands; (f) removing or digesting the hybrid **duplexes**; and (g) producing test-specific **duplexes** from the unhybridized low-abundance enriched test polynucleotide strands, is new. INDEPENDENT CLAIMS are also included for the following: (1) functionally isolating (M2) **single-stranded** polynucleotides in a mixture of single- and double-stranded polynucleotides; (2) a number of polynucleotides (I) prepared by M1, comprises at least 10 to the power of 3 different polynucleotides and is substantially enriched in sequences that are either not present in the reference polynucleotide sample or are present in the reference polynucleotide sample in substantially lower concentration than in the test polynucleotide sample, and low-abundance sequences, relative to the test polynucleotide sample; (3) a kit comprising: (a) an antisense primer or antisense primer complex comprising a sequence that binds to a primer site in the unhybridized sense test polynucleotide strands, a first restriction site 5' of the above mentioned sequence, where the first restriction site is cleaved by a restriction **endonuclease** that cleaves double-stranded polynucleotides, but leaves **single-stranded** polynucleotides substantially intact, and a first universal primer site 5' of the restriction site of above mentioned sequence; and (b) instructions for performing M1; (4) a kit comprising (I), an antisense primer complex comprising an antisense primer operably linked to an RNA promoter sequence, where the RNA promoter sequence is 5' of the antisense primer, and a sense primer, or comprising (I), and an RNA polymerase capable of transcribing antisense RNA from (I); (5) preparing (M3) a selected polynucleotide pool from a polynucleotide sample; (6) a number of polynucleotides (II) prepared from a polynucleotide sample, comprising at least 10 to the power of 3 different polynucleotides and is substantially enriched in high-abundance polynucleotide sequences relative to the polynucleotide sample, where the polynucleotides each comprise a RNA promoter sequence and a universal primer site, or is substantially enriched in low-abundance polynucleotide sequences relative to the polynucleotide sample; (7) a kit comprising an antisense primer complex which comprises an antisense primer operably linked to an RNA promoter sequence, where the RNA promoter sequence is 5' of the antisense primer, a sense primer, and instructions for performing M3; and (8) a kit comprising (II), and an RNA polymerase capable of transcribing antisense RNA from (II), or comprising (II), an antisense primer complex which comprises an antisense primer operably linked to an RNA promoter sequence, and a sense primer.

BIOTECHNOLOGY - Preferred Method: In M1, the high-abundance enriched

polynucleotide strands comprise high-abundance enriched antisense polynucleotide strands, the test polynucleotide strands comprise sense test polynucleotide strands, the low-abundance enriched test polynucleotide strands comprise antisense test polynucleotide strands, and the low-abundance enriched reference polynucleotide strands comprise enriched reference polynucleotide strands. The antisense test polynucleotide strands are synthesized from the unhybridized sense test polynucleotide strands using a first antisense primer and the first universal primer comprises a 2nd restriction site different from the first. The sense reference polynucleotide strands comprise a 3rd restriction site previously added to the 5' ends of the sense reference polynucleotide strands, where the site is cleaved by a restriction **endonuclease** that cleaves double stranded polynucleotides only. The hybrid **duplexes** are digested with a restriction **endonuclease**. The test specific **duplexes** are produced from unhybridized antisense test polynucleotide strands by amplification using a first universal primer and a second universal primer. The amplification is performed by an enhanced polymerase chain reaction. The molar ratio of the high/low abundance enriched polynucleotide strands to the test polynucleotide strands in the first hybridization mixture is 1 to 100:1. Antisense test polynucleotide strands are primed using oligonucleotide-dT priming. The test specific **duplexes** are cloned into a vector. M1 additionally comprises producing a polynucleotide **library** from the **duplexes**. An expression vector was introduced into a host cell and expressing the protein encoded by the cloned test specific **duplex**. The cell is in vitro. At least one of the **duplexes** is labeled with a detectable label. The test and reference polynucleotide samples are mRNA from a normal cell or tissue and from a diseased cell or tissue.

USE - M1 is useful for identifying one or more polynucleotides in a test sample that are absent from, or less abundant in, a reference sample (claimed). M1 is useful for identifying low-abundance sequences that are differentially expressed between different samples. (I) or (II) is useful for cloning, expression, or hybridization studies. A selected polynucleotide pool prepared by M3 is useful in nucleotide synthesis, hybridization, as driver in subtractive hybridization, to produce a nucleotide array, and in research and therapeutic uses.

EXAMPLE - Subtractive hybridization for identifying low-abundance polynucleotides that differed between a test and reference sample was as follows. An antisense test RNA pool enriched in high-abundance sequences was prepared. A pool of sense reference cDNA strands that was enriched in low-abundance sequences was also prepared. The antisense test RNA was used as driver in a first subtractive hybridization reaction with test mRNA to block the high-abundance test mRNA molecules, leaving low-abundance test mRNA molecules free to serve as a template for first-strand cDNA synthesis. Low-abundance enriched first-strand (antisense) test cDNA was synthesized from the unhybridized test mRNA molecules using reverse transcriptase and an antisense primer including an oligo-dT sequence and two restriction sites 5' of the oligo-dT sequence. One restriction site (e.g., Alu I) was cleaved by an enzyme specific for double-stranded **DNA**. The other restriction site was used to clone test-specific polynucleotides into a cloning vector after the second hybridization reaction. The reaction mixture was then treated with 100 mM NaOH at 68 degrees C for 60 minute to degrade the test mRNA, leaving low-abundance enriched antisense test cDNA strands. The sense reference cDNA of was used as driver in a subtractive hybridization reaction with the antisense test cDNA strands of to block antisense test cDNA strands corresponding to polynucleotide sequences that were present in both the test and reference samples. This second hybridization reaction produced double-stranded cDNA molecules, unhybridized low-abundance enriched antisense test cDNA strands, and unhybridized low-abundance enriched sense reference cDNA strands (i.e., the driver). The hybrid **duplexes** corresponded to sequences

present in both the test and reference sample, whereas the unhybridized antisense test cDNA strands corresponded to test-specific sequences. The sticky ends of the hybrid **duplexes** were filled in using Taq polymerase to generate double-stranded Alu I restriction sites. Oligonucleotide tailing was carried out with dC to add a poly-C tail to all molecules in the reaction mixture. The hybrid **duplexes** were digested with Alu I, which cleaved off the 3' poly-C tails at either end of the **duplex**. This reaction left intact the unhybridized low-abundance enriched antisense test cDNA strands, and unhybridized low-abundance enriched sense reference cDNA strands (i.e., the driver). Low-abundance enriched antisense test cDNA strands were converted to double-stranded test cDNA molecules by selective polymerase chain reaction (PCR) using suitable primers. The resulting mixture was digested with Sfi I and Not I and ligated into a suitable vector to produce a **library** of low-abundance enriched, test-specific clones. (99 pages)

L122 ANSWER 19 OF 42 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI
ACCESSION NUMBER: 1998-01293 BIOTECHDS
TITLE: Directional cloning of **DNA** using oligonucleotide that hybridizes to **single-stranded** cDNA; for 5'-end cloning and cDNA **library** construction
AUTHOR: Eberwine J H; Madison R
PATENT ASSIGNEE: Univ.Pennsylvania
LOCATION: Philadelphia, PA, USA.
PATENT INFO: WO 9741249 6 Nov 1997
APPLICATION INFO: WO 1997-US6957 25 Apr 1997
PRIORITY INFO: US 1996-16617 1 May 1996
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 1997-549747 [50]
AB A new improved method for directional cloning of **DNA** involves annealing a 1st oligonucleotide (ON) encoding a restriction site to ss negative-sense (-)-cDNA, to create a ds region on the cDNA, so that regions to be replicated during formation of the 2nd positive-sense (+)-strand are limited. Poly-A mRNA is primed with oligo-dT or a random **DNA** primer for (-)-strand formation, and products are given dG or dC homopolymer tails, forming **heteroduplexes**. These are denatured, the RNA component is removed using RNA-ase or alkaline hydrolysis, and the resulting cDNA is combined with the 1st ON. The ds **DNA** product is cleaved with a restriction **endonuclease**, to give ss **DNA** with a 5'-sticky end and a 3'-poly-dG/C region. A 2nd ON, complementary to the 3'-end and with the same restriction site, is annealed to the 3'-end, so that the 5'-end loops back and anneals to ON2 at the restriction site forming a primed and gapped ss (-)-cDNA. The method may be used to clone 5'-ends of **DNA**. The process increases the efficiency of cloning of 5'-ends, increasing the chance of creating full-length cDNA **libraries** from overlapping subsequences. (16pp)

L122 ANSWER 20 OF 42 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI
ACCESSION NUMBER: 1996-09751 BIOTECHDS
TITLE: Subtractive cDNA hybridization method; using nuclease-resistant tester cDNA **library** for use in differentially expressed gene isolation
AUTHOR: Zeng J
PATENT ASSIGNEE: U.S.Dep.Health-Hum.Serv.
LOCATION: Washington, DC, USA.
PATENT INFO: US 5525471 11 Jun 1996
APPLICATION INFO: US 1994-322075 12 Oct 1994
PRIORITY INFO: US 1994-322075 12 Oct 1994
DOCUMENT TYPE: Patent
LANGUAGE: English

OTHER SOURCE: WPI: 1996-286394 [29]

AB A new method for performing subtractive cDNA hybridization to obtain a 1st **library** enriched in cDNA that is not present in a 2nd **library** involves: providing a **library** of tester cDNA, where the tester cDNA is protected from digestion by a 1st nuclease (**exonuclease-III**); contacting the tester cDNA in denatured form with a **library** of denatured driver cDNA, where the driver cDNA is not protected from digestion by the 1st nuclease to form a denatured mixture; permitting cDNA in the denatured mixture to form ds cDNA comprising homoduplexes and **heteroduplexes**; digesting unprotected cDNA with the 1st nuclease; and treating the resulting material with a 2nd nuclease (exonuclease-VII) to digest ss cDNA and thereby provide a **library** enriched in tester cDNA that is not present in the driver cDNA **library**. The tester cDNA is protected from digestion by the 1st nuclease by incorporating exonuclease-resistant nucleotide analogs (deoxynucleoside thiotriphosphates) into the tester cDNA using **DNA**-polymerase (EC-2.7.7.7). Also new is a kit for performing the new method. The method is useful for differentially expressed gene isolation, etc. (9pp)

L122 ANSWER 21 OF 42 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI
ACCESSION NUMBER: 1992-12329 BIOTECHDS
TITLE: Method for coincident sequence isolation from **DNA** mixture;
useful in linkage studies, **library** analysis, expression studies and analysis of complex **DNA** mixtures
PATENT ASSIGNEE: Med.Res.Counc.
PATENT INFO: WO 9213100 6 Aug 1992
APPLICATION INFO: WO 1992-GB144 24 Jan 1992
PRIORITY INFO: GB 1991-1757 26 Jan 1991
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 1992-284682 [34]

AB A method for recovering **DNA** sequences which are homologous to those in a 2nd **DNA** mixture is new. The method comprises: using restriction **endonucleases** (REs) to produce **single-stranded** (ss) **DNA** fragments with added defined flanking sequences; annealing capture oligonucleotides (COs) to the added defined flanking sequences of the resulting ss **DNA** fragments; treating **DNA** in a 2nd mixture with the same REs to produce ss **DNA** fragments; combining the products of the 1st and 2nd mixtures; and allowing ss **DNA** with 100% homology to anneal; joining the annealed ss **DNA** from the 2nd mixture present as a **heteroduplex** to the COs from the 1st mixture; and recovering (by the polymerase chain reaction, PCR) the sequences captured in the form of **heteroduplex** coincident **DNA** including the COs. **DNA** from the 1st mixture is made ss by cloning in phage M13 or by PCR. COs are preferably 30-40 bases long. An oligonucleotide complementary to the CO is added to the products of the 1st **DNA** mixture before combining the products of the 1st and 2nd **DNA** mixtures. Highly conserved sequences of different species and within the same species may be isolated. (21pp)

L122 ANSWER 22 OF 42 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2002:778193 CAPLUS
DOCUMENT NUMBER: 137:289897
TITLE: Use of generic oligonucleotide microchips to detect protein-nucleic acid interactions in massive parallel analysis
INVENTOR(S): Krylov, Alexander; Mirzabekov, Andrei; Prokopenko, Dmitry; Rouviere-Yaniv, Josette; Zasedateleva, Olga
PATENT ASSIGNEE(S): University of Chicago, USA

SOURCE: PCT Int. Appl., 34 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2002079488	A2	20021010	WO 2001-US49555	20011227

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

PRIORITY APPLN. INFO.: US 2000-258824P P 20001228

AB Nucleic acids or proteins immobilized in a gel pad are interacting with a protein and the nucleic acid-protein and protein-protein interactions are characterized and measured. Large-scale, parallel measurements of these interactions can be examd. to provide a powerful tool in elucidating interactions between proteins and nucleic acids. The method is demonstrated by using a generic hexadeoxyribonucleotide gel-pad microchip to test the DNA-binding properties of HU histone-like bacterial protein, which is known to have a low sequence specificity. Large scale set of 8-mers oligonucleotides contg. 6-mers core with degenerate bases at both the 3'- and 5'-ends are immobilized within polyacrylamide gel pads of the microchip. The double-stranded duplexes are formed from single-stranded immobilized oligonucleotides by hybridization with a specified mixt. of 8-mers. The melting temp. (Tm) curves of ssDNA or dsDNA microchips in the presence or absence of HU protein are analyzed. The statistical data suggest HU protein forms two classes of complexes with DNA, a major one with dsDNA and a minor one with ssDNA. The major class of complexes is formed with dsDNA, which is not specific with some preferred motifs, such as AA, AAG, or GAA, that increase the Tm. The minor complexes are formed with low melting oligonucleotides and the binding decreases the Tm. Duplexes with different A/T content have different properties both for shifts of Tm and for quenching of fluorescent signals, when in complexes with HU. The results obtained support the model that in the case of the A/T-rich duplexes, HU protein binds to each single strand of dsDNA, therefore, decreasing the Tm and quenching the fluorescent signal from this gel pad. HU protein does not have a strong binding specificity for ssDNA fragments, but the binding const. is higher in the case of G/C-rich sequences. The results demonstrate that generic microchips could be an efficient approach in anal. of sequence specificity of proteins.

L122 ANSWER 23 OF 42 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2002:293883 CAPLUS
DOCUMENT NUMBER: 136:320310
TITLE: Identification and isolation of polynucleotides containing nucleic acid differences by single-stranded trap
INVENTOR(S): Thill, Gilbert
PATENT ASSIGNEE(S): Genset, Fr.
SOURCE: PCT Int. Appl., 76 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2002031190	A2	20020418	WO 2001-IB1464	20010619

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

US 2002055109	A1	20020509	US 2001-882608	20010615
AU 2001076632	A5	20020422	AU 2001-76632	20010619

PRIORITY APPLN. INFO.: US 2000-240262P P 20001012
WO 2001-IB1464 W 20010619

AB The present invention is directed to methods of isolation of related polynucleotides harboring nucleic acid difference within a polynucleotide sample. The method will be useful in detecting and identifying alternative splicing events and corresponding splicing isoforms and genomic DNA differences between genomes. The method according to the present invention is based on the use of a single-stranded trap. The single-stranded trap preferably involves the use of single-strand binding protein, such as SSB protein from E. coli. The invention can be used to identify insertion, deletion, replacement of at least 6 nucleotides. The invention can be used to identify sequence differences between the genome of two strains of given pathogen that differ in their sensitivity to a given drug.

L122 ANSWER 24 OF 42 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2002:965022 CAPLUS
DOCUMENT NUMBER: 138:20463
TITLE: Polymerase chain reaction of DNA of which base sequence is completely unidentified
INVENTOR(S): Park, Han Oh; Weon, Se-Yeon; Rhee, Joo-Won; Joung, In-Suk; Song, Su-Nam; Jeon, Jin-Tae
PATENT ASSIGNEE(S): S. Korea
SOURCE: U.S. Pat. Appl. Publ., 8 pp.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 2002192769	A1	20021219	US 2001-849597	20010507

PRIORITY APPLN. INFO.: US 2001-849597 20010507

AB The present invention relates to a process for amplifying DNA of an organism. More particularly, the present invention is directed to a process for amplifying DNA of an organism through polymerase chain reaction(PCR) without any information regarding a primer needed for amplifying DNA of an organism.

IT **9037-44-9, Exonuclease III**
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(polymerase chain reaction of DNA of which base sequence is completely unidentified)

L122 ANSWER 25 OF 42 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2002:450247 CAPLUS
DOCUMENT NUMBER: 137:16493
TITLE: Method and kits for simultaneous detection of single

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                              nucleotide polymorphisms for cancer diagnosis
INVENTOR(S):                  Platica, Ovidiu
PATENT ASSIGNEE(S):           USA
SOURCE:                       U.S. Pat. Appl. Publ., 14 pp.
                              CODEN: USXXCO
DOCUMENT TYPE:                Patent
LANGUAGE:                     English
FAMILY ACC. NUM. COUNT:       1
PATENT INFORMATION:

     PATENT NO.       KIND  DATE           APPLICATION NO.  DATE
     ---------------  ----  --------       ---------------  --------
     US 2002072057     A1   20020613       US 2001-808504   20010314
PRIORITY APPLN. INFO.:                  US 2000-190879P  P 20000321
                                        US 2001-266191P  P 20010202
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AB This invention relates to kits and methods of identifying polymorphisms assocd. with particular cancers. It involves contacting the nucleic acid of interest with a suitable ref. nucleic acid under suitable conditions that the nucleic acid of interest forms a heteroduplex with the ref. nucleic acid. Preferably, the nucleic acid of interest is RNA expressed from a cDNA library and the ref. nucleic acid is DNA or a circular nucleic acid. The heteroduplex is contacted with a combination of suitable nucleases to selectively cleave the heteroduplex if a base change(s) is present. The nucleases may be S1 nuclease or RNAse I and S1 nuclease. A labeled probe is ligated to the cleaved heteroduplex and detected under suitable conditions so as det. the location and the sequence of the base changes.

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L122 ANSWER 26 OF 42  CAPLUS  COPYRIGHT 2003 ACS
ACCESSION NUMBER:             2002:533973  CAPLUS
DOCUMENT NUMBER:              137:89425
TITLE:                        Multiplex ValiGeneSM Gene Identification (VGID-SM) for
                              identifying genes underlying a defined phenotype and
                              uses thereof
INVENTOR(S):                  Iris, Francois J. M.; Pourny, Jean-louis
PATENT ASSIGNEE(S):           Valigen (US), Inc., USA
SOURCE:                       U.S., 60 pp., Cont.-in-part of U. S. 6,221,585.
                              CODEN: USXXAM
DOCUMENT TYPE:                Patent
LANGUAGE:                     English
FAMILY ACC. NUM. COUNT:       2
PATENT INFORMATION:

     PATENT NO.       KIND  DATE           APPLICATION NO.  DATE
     ---------------  ----  --------       ---------------  --------
     US 6420111        B1   20020716       US 1999-232074   19990115
     US 6221585        B1   20010424       US 1998-7905     19980115
PRIORITY APPLN. INFO.:                  US 1998-7905     A2 19980115
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AB The present invention relates generally to the field of genomics. More particularly, the present invention relates to a method for gene identification beginning with user-selected input phenotypes. The method is referred to generally as the ValiGeneSM Gene Identification method, or the VGIDSM method. When more than two source populations of nucleic acids are simultaneously compared, the method may be referred to as multiplex VGIDSM . Multiplex VGID, also known as ValiGene Gene Trapping (VGGT), offers an alternative approach for anal. of complex, multistage systems. The method employs nucleic acid mismatch binding protein chromatog. to effect a mol. comparison of one phenotype with others. Genes are identified as having a specified function, or as causing or contributing to the cause or pathogenesis of a specified disease, or as assocd. with a specific phenotype, by virtue of their selection by the method. Identified genes may be used in development of reagents, drugs and/or combinations thereof useful in clin. or other settings for prognosis,

diagnosis and/or treatment of diseases, disorders and/or conditions. The method is equally suited for gene identification for agricultural, bio-engineering, medical, veterinary, and many other applications. The VGID method was demonstrated by the identification of the cDNA of human DinP gene, encoding a sequence homolog of the bacterial DinP protein, assocd. with the DNA damage repair pathway. The multiplex VGID (VGGT) method was demonstrated by the study of breast cancer, as an example of a complex system, using 4 cell lines of different cancer stages.

REFERENCE COUNT: 100 THERE ARE 100 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L122 ANSWER 27 OF 42 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2002:866722 CAPLUS
DOCUMENT NUMBER: 137:364392
TITLE: Selective polymerase chain reaction of DNA of which base sequence is completely unknown
INVENTOR(S): Park, Han-Oh; Weon, Se-Yoon; Rhee, Joo-Won; Joung, In-Suk; Song, Su-Nam; Jeon, Jin-Tae
PATENT ASSIGNEE(S): Bioneer Corporation, S. Korea
SOURCE: Eur. Pat. Appl., 10 pp.
CODEN: EPXXDW
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
EP 1256630	A2	20021113	EP 2002-10053	20020506
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
JP 2003009864	A2	20030114	JP 2002-131307	20020507

PRIORITY APPLN. INFO.: KR 2001-25637 A 20010507

AB The present invention relates to a process for amplifying DNA of an organism. More particularly, the present invention is directed to a process for amplifying DNA of an organism through Polymerase Chain Reaction(PCR) without any information regarding to a primer needed for amplifying DNA of an organism.

IT **9037-44-9, Exonuclease III**
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(selective polymerase chain reaction of DNA of which base sequence is completely unknown)

L122 ANSWER 28 OF 42 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2001:449218 CAPLUS
DOCUMENT NUMBER: 135:56874
TITLE: Method of generating unidirectional nested deletions using bacteriophage f1 **endonuclease**
INVENTOR(S): Dunn, John J.; Quesada, Mark A.; Randesi, Matthew
PATENT ASSIGNEE(S): Brookhaven Science Associates, USA
SOURCE: U.S., 23 pp., Cont.-in-part of U.S. 5,928,908.
CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 6248569	B1	20010619	US 1999-342353	19990629
US 5928908	A	19990727	US 1997-966958	19971110

PRIORITY APPLN. INFO.: US 1997-966958 A2 19971110

AB A method for the introduction of unidirectional deletions in a cloned DNA is described. More specifically, the method involves cloning the sequence of interest downstream of a recognition sequence for the endonuclease of bacteriophage f1, the bacteriophage replication of origin. The construct is then incubated with protein pII encoded by gene II of phage f1 to generate a single-stranded nick. The nicked DNA is then contacted with E. coli exonuclease III to convert the single-stranded nick into a single-stranded gap. The single-stranded gapped DNA is then incubated with a single-strand-specific endonuclease thereby producing a linearized DNA mol. contg. a double-stranded deletion corresponding in size to the single-stranded gap. The digestion products can then be ligated into a suitable vector. A method for producing single-stranded DNA probes is also described. Single-stranded gapped DNA produced as described above is incubated with a DNA polymerase in the presence of labeled nucleotides to fill in the gap. This DNA is then linearized by digestion with a restriction enzyme which cuts outside the DNA segment of interest. The product of this digestion is then denatured to produce a labeled single-stranded nucleic acid probe. These methods are useful for introducing deletions into either or both ends of a cloned DNA insert, for high throughput sequencing of any DNA of interest.

IT **9037-44-9, Exonuclease III**
RL: BUU (Biological use, unclassified); CAT (Catalyst use); BIOL (Biological study); USES (Uses)
(method of generating unidirectional nested deletions using bacteriophage f1 **endonuclease**)

REFERENCE COUNT: 7 THERE ARE 7 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L122 ANSWER 29 OF 42 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2001:128143 CAPLUS
DOCUMENT NUMBER: 134:188962
TITLE: mRNA 3' and 5' untranslated region cDNA **libraries** for identifying regulatory UTR sequences
INVENTOR(S): Giordano, Tony; Powers, Gordon D.; Eder, Paul S.; Barnett, Bonnie Ann; Temeles, Gretchen L.
PATENT ASSIGNEE(S): Message Pharmaceuticals, Inc., USA
SOURCE: Jpn. Kokai Tokkyo Koho, 13 pp.
CODEN: JKXXAF
DOCUMENT TYPE: Patent
LANGUAGE: Japanese
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 2001046086	A2	20010220	JP 2000-201516	20000703
US 6448007	B1	20020910	US 2000-603522	20000623
EP 1176196	A1	20020130	EP 2000-115854	20000724

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO

PRIORITY APPLN. INFO.: US 1999-142217P P 19990702

AB CDNA libraries corresponding to the mRNA 3' and 5' untranslated region (UTR) sequences, vectors contg. them, and use in identifying regulatory UTR sequences, are disclosed. Prepn. of poly A(+) RNA from whole RNA, non-random enzymic digestion of poly A(+) RNA with RNAse H, synthesis of heteroduplex contg. 5' UTR sequences, and cDNA synthesis from the heteroduplex is described. Binding of ribosome, RT-PCR with oligo(dT) primer and polymerase in cDNA library construction is also claimed. Use of a genetic construct contg. reporter genes operably linked to a promoter, one linked also to the UTR sequences and other not linked in identifying regulatory UTR sequences, is also described. A fluorescent protein or a cell surface marker can be used as reporter. Screening of

cells transformed with the construct by FACS is described.

L122 ANSWER 30 OF 42 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2001:379503 CAPLUS
DOCUMENT NUMBER: 135:16331
TITLE: **Heteroduplex** mobility assay (HMA) pre-screening: an improved strategy for the rapid identification of inserts selected from phage-displayed peptide **libraries**
AUTHOR(S): Fack, Fred; Deroo, Sabrina; Kreis, Stephanie; Muller, Claude P.
CORPORATE SOURCE: Laboratoire National de Sante, Luxembourg, L-1101, Luxembourg
SOURCE: Molecular Diversity (2001), Volume Date 2000, 5(1), 7-12
CODEN: MODIF4; ISSN: 1381-1991
PUBLISHER: Kluwer Academic Publishers
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Phage-displayed peptide libraries represent an efficient tool to isolate peptides that bind a given target mol. After several selection rounds, generally a large pool of target binding phages is obtained. Conventional anal. of the selected phage population involves extensive sequencing of many clones, most of which can be identical. We have adapted the Heteroduplex Mobility Assay (HMA) for pre-screening of phage inserts that were amplified by direct colony PCR of ELISA-pos. clones. This strategy allowed for the rapid and reproducible assignment of insert sequences to different "heteroduplex migration groups". Sequence anal. of only one representative of each HMA migration group then completes the characterization of the binding phage population. In our model expts., only 16% of HMA pre-screened clones required further sequence anal.

REFERENCE COUNT: 19 THERE ARE 19 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L122 ANSWER 31 OF 42 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2000:291294 CAPLUS
DOCUMENT NUMBER: 132:318602
TITLE: Cloning of human genes associated with a specific phenotype using banks enriched for polymorphic sequences
INVENTOR(S): Johsson, Jon J.; Weissman, Sherman M.
PATENT ASSIGNEE(S): Yale University, USA
SOURCE: PCT Int. Appl., 45 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2000024935	A2	20000504	WO 1999-US24984	19991026
WO 2000024935	A3	20000713		

W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

CA 2316436	AA	20000504	CA 1999-2316436	19991026

EP 1042512 A2 20001011 EP 1999-956666 19991026
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI
US 6506562 B1 20030114 US 1999-427104 19991026
PRIORITY APPLN. INFO.: US 1998-105667P P 19981026
WO 1999-US24984 W 19991026

AB A general method is described for screening cDNAs, genes or genome segments to directly isolate and characterize sequences assocd. with particular phenotypes. In the case of the human genome, a simplification of the starting material is needed, and a specific method to generate highly polymorphic genome subsets for this purpose is presented. The general screening method identifies DNA sequences contg. allele frequency differences when groups with dissimilar phenotypes are compared. The approach is based on math. principles of inequality. A change in the abundance ratio of homoduplexes of perfectly matched sequences to heteroduplexes of perfectly matched sequences, or, conversely, of mismatched homoduplexes to mismatched heteroduplexes, serves as an indicator of allele frequency difference. Gene banks from different individuals are prepd. using adaptors that allow adaptor-specific amplification. Heteroduplexes can be selectively recovered by binding them with a heteroduplex-binding protein such as mutS. Heteroduplex forming sequences can then be selectively amplified using methylated primers for the adaptors.

L122 ANSWER 32 OF 42 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2000:469928 CAPLUS
DOCUMENT NUMBER: 134:305840
TITLE: **Heteroduplex** mobility assay as a tool for predicting phylogenetic affiliation of environmental ribosomal RNA clones
AUTHOR(S): Bowyer, J.; Verrills, N.; Gillings, M. R.; Holmes, A. J.
CORPORATE SOURCE: Division of Environmental and Life Sciences, Commonwealth Key Centre for Biodiversity and Bioresources, Department of Biological Sciences, Macquarie University, Sydney, Australia
SOURCE: Journal of Microbiological Methods (2000), 41(2), 155-160
CODEN: JMIMDQ; ISSN: 0167-7012
PUBLISHER: Elsevier Science Ireland Ltd.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Heteroduplex mobility assay (HMA) of partial 16S rRNA gene fragments was tested as a tool for predicting bacterial phylogenetic relationships. Approx. 400-bp fragments were amplified from a selection of cloned environmental DNAs representing a range of sequence identities and phylogenetic relationships. Heteroduplexes between pairs of sequences were formed by mixing equal amts. of PCR products, denaturing and annealing. Annealed mixes were sepd. on 8% polyacrylamide gels and silver stained. Heteroduplexes were readily distinguished from reannealed homoduplex and unannealed fragments in all sequences where percentage identity was less than 95%. The heteroduplexes showed retarded electrophoretic migration with respect to homoduplexes. The relative retardation was strongly correlated to the percentage sequence identity between the two strands. The HMA is a useful tool for screening environmental clone libraries to systematically select clones representative of the phylogenetic diversity within the sample, or to selectively retrieve members of a particular phylogenetic group for more detailed study.
REFERENCE COUNT: 19 THERE ARE 19 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L122 ANSWER 33 OF 42 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 1999:708928 CAPLUS
DOCUMENT NUMBER: 131:332950
TITLE: Function-based gene discovery using unique oligonucleotide-tagged bar-coded vectors for clone tracking and automation in cDNA **library** screening
INVENTOR(S): Cen, Hui; Sun, Shaojian
PATENT ASSIGNEE(S): Genova Pharmaceuticals Corporation, USA
SOURCE: PCT Int. Appl., 68 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9955886	A1	19991104	WO 1999-US8823	19990421

W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
AU 9935727	A1	19991116	AU 1999-35727	19990421

PRIORITY APPLN. INFO.: US 1998-65775 19980424; WO 1999-US8823 19990421

AB The present invention relates generally to the field of genomics. More particularly, the present invention relates to methods for function-based gene discovery. Genes are identified as having or being assocd. with a specific function, as participating in a specific functional pathway, or as being a member of a specific functional group, by functional expression in one or more biol. readout assays. This invention is based, at least in part, on the recognition that the signal-to-noise ratio of a readout assay used to screen a cDNA library can be significantly enhanced by methods which localize multiple mol. copies of each unique clone into discrete regions or compartments prior to functional expression. In one embodiment, this invention provides methods for in situ transfection of a sorted library in a "bar-coded" vector to carry out expression of genes from libraries being screened in readout cells. The vector "bar code" is an oligonucleotide sequence within the vector which is unique to each individual clone of a library. The bar code enables sorting of the library in phys. space by hybridization to nucleic acid arrays which are complementary to library bar code sequences. The bar code unique to each clone together with the unique position of each complementary bar code in a nucleic acid array provides a method for direct retrieval of a gene having a function of interest in any given readout assay. Further, each unique bar code can serve as a specific primer for PCR and/or sequencing of a desired clone in a library. It is the ability to detect a biol. readout in a readout cell line which enables the user to identify genes having specific functions. It is able to directly screen mammalian cDNA libraries with an av. size of 106 clones through automation. Digestion of vectors is involved with restriction endonucleases. The methods set forth herein are suitable for application in a high throughput format for identification of genes and their functions simultaneously. Discovery of new genes and their functions permits development of diagnostics for early detection of diseases. This method permits discovery of discovery of disease-assocd. genes and is suitable for use with antisense libraries.

IT **9037-44-9, Exonuclease iii**
RL: ARG (Analytical reagent use); BSU (Biological study, unclassified);

ANST (Analytical study); BIOL (Biological study); USES (Uses)
(function-based gene discovery using unique oligonucleotide-tagged bar-coded vectors for clone tracking and automation in cDNA **library** screening)
REFERENCE COUNT: 7 THERE ARE 7 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L122 ANSWER 34 OF 42 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1999:27963 CAPLUS
DOCUMENT NUMBER: 130:91253
TITLE: Improved methods for isolating and recovering target DNA or RNA molecules having a desired nucleotide sequence
INVENTOR(S): Li, Wu-bo; Jesse, Joel; Nisson, Paul
PATENT ASSIGNEE(S): Life Technologies, Inc., USA
SOURCE: PCT Int. Appl., 57 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9859075	A1	19981230	WO 1998-US13043	19980624
W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, GW, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
AU 9881625	A1	19990104	AU 1998-81625	19980624
EP 1002126	A1	20000524	EP 1998-931513	19980624
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO				
US 6268133	B1	20010731	US 1998-103577	19980624
JP 2002506352	T2	20020226	JP 1999-504983	19980624
US 2002076708	A1	20020620	US 2001-829066	20010410

PRIORITY APPLN. INFO.: US 1997-50729P P 19970625
US 1998-103577 A1 19980624
WO 1998-US13043 W 19980624

AB Traditional methods for isolating particular target nucleic acid mols. are restricted by the abundance of the target sequence and by time-consuming steps. The present invention provides an improved method for the rapid isolation and recovery of a desired target DNA or RNA mol. (circular/linear and single-stranded/double-stranded) from a mixt. or library contg. such mols. The method involves the use of haptenylated probes and amino acid denaturants to select the desired mols. and eliminate the undesired library members from a sample. The invention employs hybridization methodol. combined with ligand sepn., DNA repair, and restriction enzyme digestion technol. to screen target nucleic acid mols.

IT **9037-44-9, Exonuclease III**
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(improved methods for isolating and recovering target DNA or RNA mols. having a desired nucleotide sequence)
REFERENCE COUNT: 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L122 ANSWER 35 OF 42 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.

ACCESSION NUMBER: 1999:78701 BIOSIS
DOCUMENT NUMBER: PREV199900078701
TITLE: Laboratory approaches toward gene identification.
AUTHOR(S): Marchuk, Douglas A. (1)
CORPORATE SOURCE: (1) Dep. Genetics, Duke Univ. Med. Cent., Durham, NC USA
SOURCE: Haines, J. L. [Editor]; Pericak-Vance, M. A. [Editor]. (1998) pp. 351-378. Approaches to gene mapping in complex human diseases.
Publisher: John Wiley and Sons, Inc. 605 Third Avenue, New York, New York 10158-0012, USA.
ISBN: 0-471-17195-6.
DOCUMENT TYPE: Book
LANGUAGE: English

L122 ANSWER 36 OF 42 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER: 1988:439300 BIOSIS
DOCUMENT NUMBER: BA86:91398
TITLE: NEW CLONING VECTORS AND TECHNIQUES FOR EASY AND RAPID RESTRICTION MAPPING.
AUTHOR(S): TARTOF K D; HOBBS C A
CORPORATE SOURCE: INST. FOR CANCER RES., 7701 BURHOLME AVE., PHILADELPHIA, PA. 19111.
SOURCE: GENE (AMST), (1988) 67 (2), 169-182.
CODEN: GENED6. ISSN: 0378-1119.
FILE SEGMENT: BA; OLD
LANGUAGE: English

AB We have modified plasmid, phage .lambda. and cosmid cloning vectors to be of general use for easily and unambiguously determining restriction maps of recombinant **DNA** molecules. Each vector is constructed so that it contains the rarely found NotI restriction site joined to a short synthetic linker sequence that is followed by a multiple cloning site. **DNA** cloned into these vectors may be restriction-mapped by either of two methods. In one technique, the cloned **DNA** is completely digested with NotI, followed by partial digestion with any other restriction enzyme. After electrophoresis and transfer to a nylon membrane, the fragments are hybridized to a labeled probe complementary to the NotI linker. In the second technique, referred to as recession hydridization detection, cloned **DNA** is digested with NotI and then briefly treated with **exonuclease III** to recess the 3' ends. After hybridizing a labeled complementary oligodeoxynucleotide to the **single-stranded** 5' end containing the linker sequence, the **DNA** is partially digested with another restriction enzyme, electrophoresed and the gel is exposed to x-ray film. With either method the size of each labeled fragment corresponds directly to the distance that a restriction site is located from the NotI linker terminus. Methods for obtaining partial restriction enzyme digests have been devised so that as many as 20 different enzymes may be conveniently mapped on a single gel in little more than a day. The vectors and techniques described may also be adapted to automated or semi-automated devices that read fragment lengths and calculate the resulting restriction map. In addition, the phage .lambda.-NotI vector described here may be particularly useful for constructing mammalian genomic **libraries** containing inserts bounded by a NotI site at one end and an MboI site at the other.

L122 ANSWER 37 OF 42 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER: 1987:209540 BIOSIS
DOCUMENT NUMBER: BA83:107170
TITLE: SEARCHING FOR POTENTIAL Z **DNA** IN GENOMIC ESCHERICHIA-COLI **DNA**.
AUTHOR(S): HOHEISEL J D; POHL F M
CORPORATE SOURCE: FAK. BIOL., UNIV. KONSTANZ, POSTFACH 5560, D-7750 KONSTANZ, FRG.

SOURCE: J MOL BIOL, (1987) 193 (3), 447-464.
CODEN: JMOBAK. ISSN: 0022-2836.
FILE SEGMENT: BA; OLD
LANGUAGE: English

AB The Clarke-Carbon **library** with Escherichia coli **DNA** cloned into plasmid ColE1 was partially screened for Z-**DNA** with the monoclonal antibody Z-D11 using the retardation of the covalently closed circular **DNA**-protein complex by nitrocellulose filters. About 85% of the plasmids tested at "natural" supercoil density bound to the filter. Together with binding studies of the iodinated antibody, one Z-**DNA** segment per about 18,000 base-pairs of E. coli **DNA** is observed. One clone containing the region around the lactose operon, pLC20-30, was studied in detail. Subcloning a partial Sau3A digest and selection with antibodies gave three different Z-forming sites. They were mapped to within about .+-. 20 base-pairs by preparing unidirectional deletion clones, selection of protein binding plasmids on nitrocellulose filters and subsequent sizing on agarose gels. The size of the Z-**DNA**-forming segments was estimated from two-dimensional gels of topoisomer mixtures. Together with results from sequencing of the plasmid **DNA** using **exonuclease III** to create **single-stranded** templates, stretches of alternating purine-pyrimidine tracts of 12 to 15 base-pairs were found to be responsible for Z-**DNA** formation. One of the sites was found in the middle of the lacZ gene, where it might be an obstacle for RNA polymerase. The methods used here should also be helpful for studying other **DNA**-protein sites, especially if they exist only in supercoiled **DNA**.

L122 ANSWER 38 OF 42 BIOSIS

ACCESSION NUMBER: 1984:274377 BIOSIS
DOCUMENT NUMBER: BA78:10857

TITLE: CONSTRUCTION OF A CLONED **LIBRARY** OF ADENOVIRUS **DNA** FRAGMENTS IN BACTERIO PHAGE M-13.

AUTHOR(S): EGGERDING F A; PIERCE W C
CORPORATE SOURCE: DEP. PATHOL., SCH. MED., UNIV. CALIFORNIA, LOS ANGELES, LOS ANGELES, CALIF. 90024.

SOURCE: J BIOL CHEM, (1983) 258 (16), 10090-10097.
CODEN: JBCHA3. ISSN: 0021-9258.
FILE SEGMENT: BA; OLD
LANGUAGE: English

AB The construction of recombinant M13 phages containing adenovirus **DNA** inserts was undertaken to provide strand-specific hybridization probes for analyses of adenovirus type 2 RNA transcripts. A **library** of molecular probes was constructed by cloning restriction **endonuclease** fragments of adenovirus types 2 and 5 **DNA** in the **duplex** replicative form **DNA** of the **single-stranded** bacteriophage vectors, M13mp7, M13mp8 and M13mp9. Adenovirus **DNA** segments from early, intermediate and late gene regions, accounting for at least 95% of the adenovirus chromosome, were cloned in both possible orientations using these M13 derivatives as vectors. **DNA** cloned into these vectors was readily obtained in a circular **single-stranded** form directly from mature phage particles. The cloned **DNA** fragments were oriented and further characterized by restriction **endonuclease** mapping and hybridization with 32P-labeled adenovirus **DNA**. The polarity and fidelity of the adenovirus **DNA** in the recombinant phages was confirmed by hybridization with labeled adenovirus 2 early and late mRNA. Restriction **endonuclease** analyses of M13 clones containing adenovirus **DNA** inserts spanning genome coordinates 31.7-56.9 indicated that the relative locations of some restriction coordinates located within this region do not correspond to the mapped restriction sites in the **DNA** of adenovirus 2. Potential uses for these M13 clones in studies of adenovirus

gene expression are discussed.

L122 ANSWER 39 OF 42 WPIDS (C) 2003 THOMSON DERWENT
ACCESSION NUMBER: 2003-039601 [03] WPIDS
DOC. NO. CPI: C2003-009343
TITLE: Novel polymorphisms of N-acetyltransferase 2 gene involved in drug metabolism and various disorders useful in therapeutics and to identify polymorphisms as a predisposition to various diseases e.g. cancer, leprosy.
DERWENT CLASS: B04 D16
INVENTOR(S): FITZGERALD, M; THOMANN, H; WALL, K
PATENT ASSIGNEE(S): (FITZ-I) FITZGERALD M; (THOM-I) THOMANN H; (WALL-I) WALL K
COUNTRY COUNT: 1
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
US 2002128215	A1	20020912	(200303)*		24

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
US 2002128215	A1 Provisional	US 2000-179876P	20000202
		US 2001-776407	20010202

PRIORITY APPLN. INFO: US 2000-179876P 20000202; US 2001-776407 20010202

AB US2002128215 A UPAB: 20030113

NOVELTY - An isolated **nucleic acid** (I) comprising at least 15 consecutive nucleotide bases including a polymorphic site in the wild type N-acetyltransferase 2 (NAT-2) gene, is new.

DETAILED DESCRIPTION - An isolated **nucleic acid** (I) comprising at least 15 consecutive nucleotide bases including a polymorphic site in the wild type N-acetyltransferase 2 (NAT-2) gene, is new.

(I) comprises at least 15 consecutive nucleotide bases including a polymorphic site chosen from C to G substitution at nucleotide -255, 403 or 51, C to T substitution at nucleotide -234, a T to A substitution at nucleotide 70, G to T substitution at nucleotide 609 and a G to A substitution at nucleotide 838 in the wild type NAT-2 gene which has a sequence (S1) of 1170 bp given in the specification.

INDEPENDENT CLAIMS are included for the following:

(1) an isolated allele specific primer (II) capable of detecting a polymorphic site of (S1);

(2) an isolated allele specific oligonucleotide probe (III) capable of detecting a polymorphic site of (S1);

(3) a diagnostic kit (IV) comprising (II) or (III);

(4) an isolated **nucleic acid** (V) comprising at least 50 consecutive **nucleic acids** of (S1) containing at least one of the polymorphic sites as above;

(5) an expression vector (VI) containing (I) or (V);

(6) a host cell (VII) containing (VI);

(7) an isolated polypeptide (VIII) comprising at least 5 consecutive amino acid bases, one or more of which are encoded by the nucleotides at a polymorphic site of (I) or its complement;

(8) an isolated polypeptide (IX) comprising at least 5 consecutive amino acid bases including a polymorphic site chosen from a Asn to substitution at amino acid position 17, a Leu to Ile substitution at position 24, a Leu to Val substitution at position 135, a Glu to Asp substitution at position 203, and a Val to Met substitution at position

280 of a sequence (S2) of 291 amino acids given in the specification;
(9) an isolated amino acid sequence (X) having 80% identity to (IX);
(10) an antibody (XI) or its fragment which binds to any of the above polypeptide sequences;
(11) an antisense oligonucleotide (XII) comprising at least 5 nucleotide bases of a polymorphic site of (I);
(12) detecting (M1) (I), by a method chosen from restriction fragment length polymorphism detection based on allele specific restriction **endonuclease** cleavage, hybridization with allele specific oligonucleotide probes, oligonucleotide arrays, allele specific polymerase chain reaction (PCR), mismatch repair detection (MRD), denaturing gradient gel electrophoresis (DGGE), **single strand** conformation polymorphism detection (SSCP), RNAase cleavage at mismatched bp chemical or cleavage of **heteroduplex DNA**, methods based on allele specific primer extension, genetic bit analysis (GBA), oligonucleotide ligation assay (OLA), allele specific ligation chain reaction (LCR), gap, radioactive and/or fluorescent **DNA** sequencing, and peptide **nucleic acid** (PNA) assays;
(13) identifying (M2) a polymorphism of (I) in a mammal, by preparing a sample of cells or tissue of the mammal, probing the tissue or cell with all or a portion of a polymorphism of (I) under conditions where hybridized **DNA** can be produced, identifying the hybridized **DNA** and cloning and sequencing the hybridized **DNA** to obtain and identify the NAT-2 gene in the mammal;
(14) treating (M3) a NAT-2 disorder by administering a molecule which binds to an endogenous analog of NAT-2 or a compound which is an agonist or antagonist of (I), its variant or fragment;
(15) labeling (M4) an individual in a clinical trial, by producing a **library** of SNPs including the polymorphic sites of (I) and their respective phenotype, sequencing an individuals NAT-2 gene, matching the genotype with the phenotype;
(16) creating (M5) a prognosis protocol by identifying patients receiving at least one NAT-2 drug, determining whether they are rapid acetylator or a slow acetylator, and converting the data obtained into a prognosis protocol;
(17) identifying (M6) therapeutic compositions which are efficacious in individuals, by administering a therapeutic composition to an individual and measuring its efficacy, determining by the individual's genotype and the polymorphic sites of (I) whether the individual is a rapid acetylator and slow acetylator, and determining which therapeutic composition will be the most effective for that particular genotype and which will have the least adverse effects;
(18) identifying (M7) an individual, by sequencing an individual's NAT-2 gene, comparing the results the frequency of NAT-2 in the population as given in the specification, using the data with other polymorphic sites in the human genome to statistically conclude the likelihood of the set of SNPs from this individual as compared to the general population;
(19) genetically linking (M8) a first individual to a second individual, by sequencing the NAT-2 gene of the first individual and parents of the second individual, comparing the particular SNPs from the two parents with the SNPs of the second individual, matching SNPs of the parents of the second individual and assessing, through statistical methods utilizing the frequency given in the specification, the likelihood of this frequency of SNPs in the general population; and
(20) a computer readable medium (XIII) comprising (I).
ACTIVITY - Cytostatic; Antileprotic. No biological data given.
MECHANISM OF ACTION - Gene therapy; Modulator of (I).
USE - (I) is useful for diagnosis and gene therapy, to identify **DNA** probes for NAT-2 genes, PCR primers to amplify NAT-2 genes and regulatory elements of the NAT-2 genes. (I) is useful for identifying individuals and in paternity testing. (I) is useful as a valuable information source to characterize individuals in terms of haplotypes and other sub-groupings, such as investigation of susceptibility to treatment

with particular drugs. The polynucleotide sequences are particularly useful as components in databases useful for sequence identity and other search analyses. (IX) is useful as an immunogen to generate antibody that binds the polymorphic protein, and for screening for drugs. (M3) is useful for treating NAT-2 disorders such as bladder cancer, colon cancer, prostate cancer, Gilbert's disease and leprosy. (I) is useful in diagnosing individuals with NAT-2 polymorphisms which are associated with these diseases and affect the metabolism of the compounds. (M5) is useful for creating a prognosis protocol for a patient receiving a therapeutic composition metabolized by NAT-2 such as isoniazid, phenylzine, hydrazine, dapsone, procainamide, sulfamethazine and other sulfonamides. The prognosis protocol includes prediction of drug efficacy, prediction of patient's prognosis, prediction of drug interaction and prediction of adverse effects. Cells and animals that carry the NAT-2 gene or its analog are useful as model systems to study and test for substances that have potential as therapeutic agents.
Dwg.0/2

L122 ANSWER 40 OF 42 WPIDS (C) 2003 THOMSON DERWENT
ACCESSION NUMBER: 2001-290916 [30] WPIDS
CROSS REFERENCE: 2001-282160 [29]
DOC. NO. CPI: C2001-089272
TITLE: Forming chimeric polynucleotides, involves hybridizing **single-stranded** polynucleotide template with random population of oligonucleotides, and treating hybridized oligonucleotides to form chimeric polynucleotide.
DERWENT CLASS: B04 D16
INVENTOR(S): ARENSDORF, J J; COCO, W M; CRIST, M J; DARZINS, A; ENCELL, L P; FOLSOM, B R; HEKTOR, H J; LEVINSON, W E; NALTY, M S; PALMER, S R
PATENT ASSIGNEE(S): (ENCH-N) ENCHIRA BIOTECHNOLOGY CORP
COUNTRY COUNT: 94
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2001029211	A2	20010426	(200130)*	EN	158
AU 2001010980	A	20010430	(200148)		

WO 2001029211:
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ NL OA PT SD SE SL SZ TZ UG ZW
W: AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CR CU CZ DE DK DM DZ EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT TZ UA UG US UZ VN YU ZA ZW

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2001029211	A2	WO 2000-US29046	20001019
AU 2001010980	A	AU 2001-10980	20001019

FILING DETAILS:

PATENT NO	KIND		PATENT NO
AU 2001010980	A	Based on	WO 200129211

PRIORITY APPLN. INFO: US 2000-618935 20000718; US 1999-160420P 19991019; US 2000-514660 20000229; US 2000-218883 20000718; US 2000-219087 20000718; US 2000-219090 20000718; US

2000-618696 20000718

AB WO 200129211 A UPAB: 20011129
NOVELTY - Forming (F1) at least one chimeric polynucleotide, involves contacting a **single-stranded** polynucleotide template with a random population of oligonucleotides, under conditions where at least two oligonucleotides hybridize to the template, and treating the hybridized oligonucleotides, such that a chimeric polynucleotide is formed.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following:

(1) generating (G1) at least one chimeric polynucleotide with one or more characteristics altered in comparison to a reference polynucleotide;

(2) a kit (K) for performing directed evolution, comprising components for fragmenting oligonucleotides, for filling-in gaps, for trimming flaps, for proofreading, for incorporating uracils in templates or for modifying templates, lambda exonuclease, DNase I, and optionally, dioxyuracil triphosphate and uracil **DNA** glycosylase;

(3) a chimeric polynucleotide (I) prepared by the above method;

(4) preparing (P1) a **single-stranded** polynucleotide template containing mRNA suitable for use in forming chimeric polynucleotide;

(5) preparing (P2) mRNA transient template in vivo for use in forming (I) by:

(a) preparing a **DNA** molecule comprising a suitable insert to be transcribed;

(b) transforming a suitable cell with the **DNA** molecule such that transcription of the **DNA** molecule occurs;

(c) lysing the transformed cell; and

(d) purifying the transcribed mRNA product from the cell lysate;

(6) preparing (P3) mRNA transient template in vitro for use in forming a chimeric polynucleotide by:

(1) preparing a **DNA** molecule comprising a suitable vector and an insert to be transcribed;

(2) transcription of the **DNA** molecule utilizing elements required for transcription; and

(3) purifying the transcribed mRNA product;

(7) a template (II) formed by the above in vitro or in vivo method;

(8) preparing (P4) a modified **single-stranded** polynucleotide template suitable for use in forming a chimeric polynucleotide with an increased degree of chimeragenesis relative to a chimeric polynucleotide formed using an unmodified **single-stranded** polynucleotide template, by obtaining a **single-stranded** polynucleotide by isolating a polynucleotide from a suitable **nucleic acid** source, synthetically manufacturing a polynucleotide or cleaving the polynucleotide from a larger polynucleotide, and amplifying the polynucleotide obtained, treating the **single-stranded** template, such that the template is modified in a manner that the step of filling the gaps is altered, thereby increasing the degree of chimeragenesis of the polynucleotide product obtained as by the above process;

(9) producing (P5) a **library** of chimeric polynucleotides by:

(a) preparing a number of **single-stranded** templates comprising a phage vector and a polynucleotide insert;

(b) contacting the **single-stranded** templates with a population of oligonucleotides, such that at least two of the oligonucleotides hybridize to more than one template; and

(c) treating the oligonucleotides hybridized to each template such that more than one contiguous chimeric polynucleotide is formed, thereby generating the **library** of chimeric polynucleotides;

(10) a **library** of chimeric polynucleotides produced by the above method;

(11) producing (P6) a **single-stranded**

polynucleotide template for forming a chimeric polynucleotide;

(12) a **single-stranded** polynucleotide produced by the above method;

(13) generating (G2) a population of **single-stranded** oligonucleotides for use in directed evolution.

(14) forming (F2) a chemically modified **single-stranded** polynucleotide template for use in directed evolution;

(15) a **single-stranded** template produced by the above method;

(16) forming (F3) a randomly fragmented oligonucleotide population for use in forming at least one chimeric polynucleotide by:

(a) treating a double-stranded polynucleotide comprising a parent polynucleotide such that a number of modified bases are formed on either or both strands; and

(b) treating the double-stranded polynucleotide such that single-stranded nicks are created as a result of the treatment and the modified bases, thus forming a randomly fragmented population of oligonucleotides for forming at least one chimeric polynucleotide;

(17) a single-stranded polynucleotide produced by the above method;

(18) forming (F4) a population of single-stranded oligonucleotides for forming at least one chimeric polynucleotide by:

(a) obtaining at least two double-stranded polynucleotides;

(b) treating the double-stranded polynucleotides with an exonuclease, thereby obtaining from each double-stranded polynucleotide, a strand containing a 3' phosphate;

(c) annealing these strands to form heteroduplex nucleic acids;

(d) treating the heteroduplex nucleic acids with an enzyme that cleaves mismatches to yield homoduplexes; and

(e) treating the homoduplexes with an enzyme to degrade the strand containing the incorporated uracils, thereby forming a population of single-stranded oligonucleotides for use in directed evolution;

(19) forming (F5) a population of oligonucleotides for use in forming at least one chimeric polynucleotide by:

(a) obtaining a double-stranded nucleic acid polynucleotide;

(b) obtaining a second double-stranded nucleic acid;

(c) denaturing both double-stranded nucleic acids thereby obtaining single-stranded polynucleotides;

(d) annealing these polynucleotides to form heteroduplex nucleic acids; and

(e) treating the heteroduplex nucleic acids with an enzyme that cleaves mismatches to yield oligonucleotides, thereby forming a population of oligonucleotides for use in forming at least one chimeric polynucleotide; and

(20) a population of single-stranded oligonucleotides produced by the above method.

USE - (II) is useful for forming chimeric oligonucleotides, by preparing a single-stranded polynucleotide template, modifying the template by chemical modification, creating abasic residues on the template, treatment with uracil glycosylase, physical modification or by hybridization of a pre-selected oligonucleotide resistant to nucleotide extension, to the template at its terminus, such that the degree of chimeragenesis is increased, contacting the modified template with a population of oligonucleotides, under conditions such that at least two of the oligonucleotides hybridize to the template, filling in gaps between the hybridized oligonucleotides on the template, and ligating adjacently hybridized oligonucleotides to form the chimeric polynucleotides (claimed). Methods of the invention are also useful in directed evolution.

Dwg.0/16

L122 ANSWER 41 OF 42 WPIDS (C) 2003 THOMSON DERWENT
ACCESSION NUMBER: 2000-594650 [56] WPIDS
CROSS REFERENCE: 1997-145245 [13]; 1997-319766 [29]; 1998-101069 [09];

1998-609243 [51]; 1999-263358 [22]; 1999-313351 [26]; 2000-524416 [47]; 2000-587434 [55]; 2001-050094 [06]; 2002-083006 [11]; 2002-393965 [42]

DOC. NO. CPI: C2000-177667

TITLE: Directed evolution method for preparing polynucleotides encoding a polypeptide, termed DirectEvolution (RTM), useful for obtaining novel enzymes that have optimized physical and/or biological properties.

DERWENT CLASS: B04 D16

INVENTOR(S): DJAVAKHISHVILI, T D; FREY, G J; SHORT, J M

PATENT ASSIGNEE(S): (DIVE-N) DIVERSA CORP

COUNTRY COUNT: 92

PATENT INFORMATION:

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       SK SL TJ TM TR TT TZ UA UG UZ VN YU ZA ZW
AU 2000040394 A  20001016 (200106)
EP 1092041    A1 20010418 (200123)  EN
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APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2000058517	A1	WO 2000-US8245	20000327
AU 2000040394	A	AU 2000-40394	20000327
EP 1092041	A1	EP 2000-919763	20000327
		WO 2000-US8245	20000327

FILING DETAILS:

PATENT NO	KIND		PATENT NO
AU 2000040394	A	Based on	WO 200058517
EP 1092041	A1	Based on	WO 200058517

PRIORITY APPLN. INFO: US 1999-332835 19990614; US 1999-276860 19990326

AB WO 200058517 A UPAB: 20021125

NOVELTY - A directed evolution method for preparing polynucleotides encoding a polypeptide, comprising generating site-directed mutagenesis optionally in combination with polynucleotide chimerization, selecting potentially desirable progeny molecules, and screening the polynucleotides, is new. The method is termed DirectEvolution (RTM).

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are included for the following:

(1) a method (M1) for producing a mutagenized progeny polynucleotide, comprising subjecting a starting or parental polynucleotide set to an in vitro exonuclease mediated reassembly process, where:

(a) the exonuclease-mediated reassembly process is exemplified by subjection to a 3' exonuclease treatment, such as treatment with **exonuclease III**, which acts on 3' underhangs and blunt ends, to liberate 3'-terminal but not 5'-terminal nucleotides from a starting double stranded polynucleotide, leaving a remaining strand that is partially or completely free of its original partner so that, if

desired, the remaining strand may be used to achieve hybridization to another partner;

(b) the exonuclease-mediated reassembly process is further exemplified by subjection to a 5' exonuclease treatment, such as treatment with red alpha gene product, that acts on 5' underhangs to liberate 5'-terminal nucleotides from a starting double stranded polynucleotide, leaving a remaining strand that is partially or completely free of its original partner so that, if desired, the remaining strand may be used to achieve hybridization to another partner;

(c) the exonuclease-mediated reassembly process is further exemplified by subjection to an exonuclease treatment, such as treatment with Mung Bean Nuclease or treatment with S1 Nuclease or treatment with Escherichia coli **DNA** polymerase, that acts on overhanging ends, including on unhybridized ends, to liberate terminal nucleotides from an unhybridized single-stranded end of an annealed **nucleic acid** strand in a **heteromeric nucleic acid** complex, leaving a shortened but hybridized end to facilitate polymerase-based extension and/or ligase-mediated ligation of the treated end; and

(d) the exonuclease-mediated reassembly process is also exemplified by a dual treatment, that can be performed, for example, non-simultaneously, with both an exonuclease that liberates terminal nucleotides from underhanging ends or blunt ends as well as an exonuclease that liberates terminal nucleotides from overhanging ends such as unhybridized ends;

(2) a method (M2) for producing a mutagenized progeny polynucleotide having at least 1 desirable property comprising:

(a) subjecting a starting or parental polynucleotide set to an in vitro exonuclease mediated reassembly process; and

(b) subjecting the progeny polynucleotide set to an end selection-based screening and enrichment process, so as to select for a desirable subset of the progeny polynucleotide set, where:

(i) the above steps can be performed iteratively and in any order and in combination;

(ii) the end selection-based process creates ligation-compatible ends;

(iii) the creation of ligation-compatible ends is optionally used to facilitate one or more intermolecular ligations, that are preferably directional ligations, within members of the progeny polynucleotide set so as to achieve assembly and/or reassembly mutagenesis;

(iv) the creation of ligation-compatible ends serves to facilitate ligation of the progeny polynucleotide set into an expression vector system and expression cloning;

(v) the expression cloning of the progeny polynucleotide set serves to generate a polypeptide set;

(vi) the generated polypeptide set can be subjected to an expression screening process; and

(vii) expression screening of the progeny polypeptide set provides a means to identify a desirable species, e.g. a mutant polypeptide or alternatively a polypeptide fragment, that has a desirable property, such as a specific enzymatic activity;

(3) a method (M3) for generating a mutagenized progeny polynucleotide from a collection of progenitor polynucleotides comprising:

(a) annealing a poly-binding **nucleic acid** strand to two mono-binding **nucleic acid** strands to generate an annealed **heteromeric** complex of **nucleic acid** strands, where:

(i) the poly-binding **nucleic acid** strand and the two mono-binding **nucleic acid** strands are each derived from a different molecular species in the collection of progenitor polynucleotides;

(ii) the collection of progenitor polynucleotides is preferably comprised of nonidentical, though possibly related, progenitor

polynucleotides, as exemplified by a collection of genes encoding dehalogenases; and

(iii) the poly-binding **nucleic acid** strand to two mono-binding **nucleic acid** strands each have at least a 7 nucleotide-long sequence of identity to the progenitor polynucleotide from which it is derived; and

(b) subjecting the unhybridized single-stranded ends of the annealed mono-binding **nucleic acid** strands in the **heteromeric** complex to an exonuclease treatment that degrades the unhybridized ends, where:

(i) the annealment of working poly-binding and mono-binding strands derived from nonidentical polynucleotides allows the generation of a chimerization of the nonidentical polynucleotide;

(ii) in a **library** of the annealed complexes of **nucleic acid** strands, many component strands have unhybridizable ends that are suboptimal or not serviceable for priming polymerase-based extension; and

(iii) the exonuclease treatment removes such unhybridizable ends to convert the annealed complexes of **nucleic acid** strands into better primers for polymerase-based extension.

USE - The methods are useful for obtaining novel enzymes that have optimized physical and/or biological properties. By using the methods, genetic vaccines, enzymes, small molecules, and other desirable molecules can be evolved towards desirable properties. For example, vaccine vectors can be obtained that exhibit increased efficacy for use as genetic vaccines. Vectors obtained using the methods can have enhanced antigen expression, increased uptake into a cell, increased stability in a cell, and the ability to tailor an immune response. The methods are also useful for obtaining a variety of novel biologically active molecules, in the fields of antibiotics, pharmacotherapeutics, and transgenic traits.

ADVANTAGE - The methods allow the reassembly of **nucleic acid** strands that would otherwise be problematic to chimerize.

Dwg.0/6

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L122 ANSWER 42 OF 42  WPIDS (C) 2003 THOMSON DERWENT
ACCESSION NUMBER:     2000-224362 [19]   WPIDS
DOC. NO. CPI:         C2000-068531
TITLE:                Producing nucleic acids lacking
                      3'-untranslated regions to optimize production of fusion
                      proteins, used to produce fusion libraries for
                      identification of protein protein interactions and drug
                      targets for the production of protein chips.
DERWENT CLASS:        B04 D16
INVENTOR(S):          HAMMOND, P W; LIPOVSEK, D
PATENT ASSIGNEE(S):   (PHYL-N) PHYLOS INC; (HAMM-I) HAMMOND P W; (LIPO-I)
                      LIPOVSEK D
COUNTRY COUNT:        87
PATENT INFORMATION:

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     AU 9954883    A  20000306 (200030)
     EP 1105516    A1 20010613 (200134)  EN
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     US 6312927    B1 20011106 (200170)
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JP 2002522091 W 20020723 (200263) 47
US 2002160377 A1 20021031 (200274)

APPLICATION DETAILS:

PATENT NO	KIND		APPLICATION	DATE
WO 2000009737	A1		WO 1999-US18603	19990816
AU 9954883	A		AU 1999-54883	19990816
EP 1105516	A1		EP 1999-941179	19990816
			WO 1999-US18603	19990816
US 6312927	B1	Provisional	US 1998-96818P	19980817
			US 1999-374962	19990816
JP 2002522091	W		WO 1999-US18603	19990816
			JP 2000-565171	19990816
US 2002160377	A1	Provisional	US 1998-96818P	19980817
		Div ex	US 1999-374962	19990816
			US 2001-910518	20010720

FILING DETAILS:

PATENT NO	KIND		PATENT NO
AU 9954883	A	Based on	WO 200009737
EP 1105516	A1	Based on	WO 200009737
JP 2002522091	W	Based on	WO 200009737
US 2002160377	A1	Div ex	US 6312927

PRIORITY APPLN. INFO: US 1998-96818P 19980817; US 1999-374962 19990816; US 2001-910518 20010720

AB WO 200009737 A UPAB: 20000419

NOVELTY - A method for removing the 3'-untranslated region of a **DNA** molecule comprising an open reading frame (ORF), comprises providing a **DNA** molecule having an ORF and a 3'-untranslated region, the **DNA** having a 5' overhang and a blunt end at the 3' end, and treating the **DNA** molecule with a 3'-5'exonuclease followed by a single-stranded nuclease to remove the 3'-untranslated region.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following:

(1) a method for removing the 3'-untranslated region of an mRNA molecule, comprising

(a) translating an mRNA molecule in vitro in a translation reaction mixture lacking functional translation release factor activity, resulting in pausing of the translation reaction mixture ribosomes at the stop codon of the mRNA molecule;

(b) adding reverse transcriptase and an oligonucleotide primer which is complementary to the 3'-untranslated region of the mRNA molecule at a site proximal to the stop codon, under conditions which allow the synthesis of a strand of **DNA** that is complementary to the 3'-untranslated region and terminates at a site proximal to the stop codon; and

(c) removing the RNA portion of the RNA-**DNA duplex** formed;

(2) a method for removing the 3'-untranslated region of an mRNA molecule comprising

(a) providing a population of mRNA molecules;

(b) synthesizing strands of **DNA**, each of which is complementary to one of the mRNA molecules, using a random primer mixture where the primers each have, a 3' region comprising a stop codon flanked by a random oligonucleotide located 3', 5', or both to the stop codon, and a 5' region comprising a Type IIS restriction site;

(c) ligating to the 3'ends of the **DNA** products of step (b)

an oligonucleotide tail;
(d) amplifying the products using a first primer which is complementary to the Type IIS restriction site-containing sequence, and a second primer which is complementary to the oligonucleotide tail; and
(e) treating the products with Type IIS restriction enzyme to cleave the products;
(3) a method for removing the 3'-untranslated regions and stop codons of a population of mRNA molecules comprising
(a) providing a population of mRNA molecules;
(b) synthesizing strands of **DNA** as in step (b) of (2) but where the primers each have a 5'region which lacks a stop codon in at least one reading frame and a random 3' region; and
(c) synthesizing strands of **DNA** complementary to the **DNA** strands of step (b) using a second random primer mixture;
(4) a method for producing an RNA-protein fusion from an mRNA having a 3'-untranslated region comprising
(a) providing the **DNA** products from the previous methods;
(b) transcribing the **DNA** to produce RNA lacking a 3'-untranslated region;
(c) covalently bonding to the RNA a peptidyl acceptor, the peptidyl acceptor being positioned 3' of the protein coding sequence of the RNA; and
(d) translating the product to produce an RNA-protein fusion;
(5) a method for producing an RNA-protein fusion from a **nucleic acid** having a 3'-untranslated region comprising
(a) providing the RNA product of methods of (1) or (2), where the RNA product lacks a 3'-untranslated region;
(b) covalently bonding to the RNA a peptidyl acceptor which is positioned 3' of the protein coding sequence of the RNA; and
(c) translating the product to produce an RNA-protein fusion;
(6) a **library** of **nucleic acid** molecules, each comprising an ORF and lacking the 3'-untranslated region normally associated with the ORF;
(7) a **library** of **nucleic acid** molecules produced by the novel method or the method of (1), (2) or (3); and
(8) an RNA-protein fusion produced by the methods of (3) and (4).
USE - The fusion **libraries** can be used for the identification of protein-protein interactions, identification of drug targets, and hybridization to solid supports to create protein chips (or beads). The RNA-protein molecules may be arranged in spatially defined arrays on the protein chips to carry out large scale screening for protein or compound identification.
Dwg.0/14

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FILE COVERS 1907 - 5 Mar 2003  VOL 138 ISS 10
FILE LAST UPDATED: 4 Mar 2003  (20030304/ED)

  This file contains CAS Registry Numbers for easy and accurate
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L11          893 SEA FILE=CAPLUS ABB=ON  HETERODUPLEX?/OBI
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    GUIDES, PLEASE VISIT:
    http://www.derwent.com/userguides/dwpi_guide.html <<<


L57        13227 SEA FILE=WPIDS ABB=ON  LIBRAR?
L58       123232 SEA FILE=WPIDS ABB=ON  HETERO?
L59        10822 SEA FILE=WPIDS ABB=ON  DUPLEX?
L60          207 SEA FILE=WPIDS ABB=ON  HETERODUPLEX?
L61        68152 SEA FILE=WPIDS ABB=ON  DNA OR ?NUCLEIC ACID#
L62         1610 SEA FILE=WPIDS ABB=ON  ENDONUCLEASE# OR ENDO NUCLEASE#
L63          159 SEA FILE=WPIDS ABB=ON  (EXO OR EXONUCLEASE#)(1W)(III OR 3)
L64         3452 SEA FILE=WPIDS ABB=ON  SINGLESTRAND? OR SINGLE STRAND?
L128        7424 SEA FILE=WPIDS ABB=ON  MICROPARTICLE# OR MICRO PARTICLE#
L130           6 SEA FILE=WPIDS ABB=ON  L57 AND L61 AND L128 AND ((L58 OR L59
                 OR L60) OR (L62 OR L63 OR L64))

=> s l130 not (l65 or l119)   previously printed

L137           4 L130 NOT (L65 OR L119)

=> fil biotechds; d que l133
```



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FILE LAST UPDATED: 24 FEB 2003     <20030224/UP>

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>>> NEW CLASSIFICATION SYSTEM FROM 2002 ONWARDS - SEE HELP CLA <<<


L74         9646 SEA FILE=BIOTECHDS ABB=ON  LIBRAR?
L75        12955 SEA FILE=BIOTECHDS ABB=ON  HETERO?
L76         1136 SEA FILE=BIOTECHDS ABB=ON  DUPLEX?
L77          343 SEA FILE=BIOTECHDS ABB=ON  HETERODUPLEX?
```

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L78        96580 SEA FILE=BIOTECHDS ABB=ON  DNA OR (NUCLEIC OR DEOXYRIBONUCLEIC)
                 (W)ACID#
L79         5000 SEA FILE=BIOTECHDS ABB=ON  ENDONUCLEASE# OR ENDO NUCLEASE#
L80          212 SEA FILE=BIOTECHDS ABB=ON  (EXO OR EXONUCLEASE#)(1W)(III OR 3)

L81         3171 SEA FILE=BIOTECHDS ABB=ON  SINGLESTRAND? OR SINGLE STRAND?
L131         300 SEA FILE=BIOTECHDS ABB=ON  MICROPARTICLE# OR MICRO PARTICLE#
L133           4 SEA FILE=BIOTECHDS ABB=ON  L74 AND L78 AND L131 AND ((L75 OR
                 L76 OR L77) OR (L79 OR L80 OR L81))


=> s l133 not (l83 or l120)
```

previously printed

```
L138           2 L133 NOT (L83 OR L120)

=> fil biosis; d que l135

FILE 'BIOSIS' ENTERED AT 10:27:38 ON 05 MAR 2003
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FILE COVERS 1969 TO DATE.
CAS REGISTRY NUMBERS AND CHEMICAL NAMES (CNs) PRESENT
FROM JANUARY 1969 TO DATE.

RECORDS LAST ADDED: 26 February 2003 (20030226/ED)



L95        45266 SEA FILE=BIOSIS ABB=ON  LIBRAR?
L96       717927 SEA FILE=BIOSIS ABB=ON  DNA OR ?NUCLEIC ACID#
L134        2619 SEA FILE=BIOSIS ABB=ON  MICROPARTICLE# OR MICRO PARTICLE#
L135           3 SEA FILE=BIOSIS ABB=ON  L95 AND L96 AND L134


=> s l135 not (l121 or l103)
```

previously printed

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L139           3 L135 NOT (L121 OR L103)

=> dup rem l138,l139,l137
FILE 'BIOTECHDS' ENTERED AT 10:28:00 ON 05 MAR 2003
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FILE 'BIOSIS' ENTERED AT 10:28:00 ON 05 MAR 2003
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FILE 'WPIDS' ENTERED AT 10:28:00 ON 05 MAR 2003
COPYRIGHT (C) 2003 THOMSON DERWENT
PROCESSING COMPLETED FOR L138
PROCESSING COMPLETED FOR L139
PROCESSING COMPLETED FOR L137
L140           9 DUP REM L138 L139 L137 (0 DUPLICATES REMOVED)
                 ANSWERS '1-2' FROM FILE BIOTECHDS
                 ANSWERS '3-5' FROM FILE BIOSIS
                 ANSWERS '6-9' FROM FILE WPIDS

=> d ibib ab 1-9; fil hom

L140  ANSWER 1 OF 9  BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI
ACCESSION NUMBER: 2002-19661  BIOTECHDS
TITLE:            New biocompatible and biodegradable polymer compounds for
                  preparing drug delivery device, pharmaceutical composition,
                  polynucleotide/polymer complex;
                     complex containing polymer and encapsulated DNA
```

useful for tissue engineering, prophylaxis therapy, gene therapy and diagnosis

AUTHOR: LANGER R S; LYNN D M; PUTNAM D; AMIJI M M; ANDERSON D G
PATENT ASSIGNEE: MASSACHUSETTS INST TECHNOLOGY
PATENT INFO: WO 2002031025 18 Apr 2002
APPLICATION INFO: WO 2000-US31270 10 Oct 2000
PRIORITY INFO: US 2001-969431 2 Oct 2001
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2002-557381 [59]

AB DERWENT ABSTRACT:

NOVELTY - Polymer compounds (I)-(VIII) are new.

DETAILED DESCRIPTION - Polymer compounds of formulae (I)-(IV) and (V)-(VIII) and their derivatives and salts are new. A1, B1 = linker of 1-30C carbon chain or 1-30C **heteroatom** containing carbon chain, (substituted by at least one of alkyl, alkenyl, alkynyl, amino, alkylamino, dialkylamino, trialkylamino, aryl, ureido, (aromatic) **heterocyclic**, (aromatic) cyclic, halogen, hydroxyl, alkoxy, cyano, amide, carbamoyl, carboxylic acid, ester, carbonyl, carbonyldioxyl, alkylthioether or thiol); or L1 = absent, single bond or a linker group additionally bonded to A1 to form a cyclic structure; R1-R10 = H, alkyl, alkenyl, alkynyl, aryl, halogen, hydroxyl, alkoxy, carbamoyl, carboxyl ester, carbonyldioxyl, amide, thiohydroxyl, alkylthioether, amino, alkylamino, dialkylamino, trialkylamino, cyano, ureido, substituted alkanoyl, cyclic, aromatic cyclic, **heterocyclic**, aromatic **heterocyclic** (all optionally substituted by alkyl, alkenyl, alkynyl, amino, alkylamino, dialkylamino, trialkylamino, aryl, ureido, (aromatic) **heterocyclic**, (aromatic) cyclic, halogen, hydroxyl, alkoxy, cyano, amide, carbamoyl, carboxylic acid, ester, carbonyl, carbonyldioxyl, alkylthioether or thiol); M1 = 1,3-imidazolidinyl or a group of formula (a)-(c); M' = (CH2)2CH(CH3) or (CH2)4; S1 = 4-hydroxybutyl, 2-(2-hydroxyethoxy)ethyl, 3-(dimethylamino)propyl or 3-morpholinopropyl; X- = chloride, fluoride, bromide, iodide, sulfate, nitrate, fumarate, acetate, carbonate, stearate, laurate or oleate; n = 5-10000; m = 0-50; and a = 3-10000. INDEPENDENT CLAIMS are included for the following: (1) composition comprising polynucleotide and polymer compound; (2) synthesizing poly(beta-amino ester) (PBAE) by reacting bis-(secondary amine) and bis(acrylate ester under suitable conditions; (3) encapsulating an agent in poly(beta-amino ester) matrix to form **microparticles** by contacting agent and PBAE under suitable condition to form **microparticles**; and (4) screening a **library** of polymers which involves providing several polymers and screening polymers for desired property useful in gene therapy.

USE - For preparing drug delivery device, pharmaceutical composition, nanometer scale complexes with **nucleic acid**, polynucleotide/polymer complexes, encapsulation of therapeutic diagnostic and/or prophylactic agent e.g. polynucleotides to form **microparticles**.

ADMINISTRATION - May be administered orally, rectally, parenterally, intracisternally, intravaginally, intranasally, intraperitoneally, topically, buccally or as oral or nasal spray.

ADVANTAGE - Polymer is biodegradable and biocompatible.

EXAMPLE - 1,4-butanediol diacrylate (0.750 g) and diamine (selected from N',N'-dimethylethylenediamine, piperazine and 4,4'-trimethylenedipiperidine, 3.78 mmol) were weighted separately and dissolved in 5 mm THF. Solution containing diamine was added to diacrylate solution. The reaction was sealed and heated at 50 degrees C. After 48 hours, the reaction was cooled and slowly into vigorously stirring diethyl ether or hexanes. Polymers (poly-1, poly-2, poly-3) obtained was collected and dried under vacuum. Structure of polymers were confirmed by 1H NMR and 13C NMR which confirmed that polymers were formed through conjugate addition of secondary amines to acrylate moieties of

1,4-butanediol diacrylate. Newly formed tertiary amines in polymer backbone did not participate in subsequent addition with diacrylate monomer. Polymers were soluble inorganic solvent and water at reduced pH. Polymer (1) and hydrochloride salts of polymers (1)-(3) were freely soluble in water. Polymers had ability to complex plasmid **DNA** through agarose gel shift assay, compact plasmid **DNA** into nanometer sized structures. Polymer 1 form complexes with diameter 90-150 at **DNA**/polymer ratio 1:1. Polymer 2 condensed **DNA** into particles of 60-125 nm at **DNA**/ polymer ratio of 1:10. Polymer 3 formed complexes with diameter 50-150 nm at **DNA**/ polymer ratio of 1:2. (133 pages)

L140 ANSWER 2 OF 9 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI
ACCESSION NUMBER: 2002-10448 BIOTECHDS
TITLE: Ovarian tumor polypeptide and polynucleotide useful in diagnosis, prevention and/or treatment of cancer, especially ovarian cancer;
vaccinia virus vector and liposome-mediated recombinant protein gene transfer and expression in host cell for ovary cancer diagnosis and gene therapy
AUTHOR: XU J; STOLK J A; ALGATE P A; FLING S P
PATENT ASSIGNEE: XU J; STOLK J A; ALGATE P A; FLING S P
PATENT INFO: US 2002004491 10 Jan 2002
APPLICATION INFO: US 1999-825294 10 Sep 1999
PRIORITY INFO: US 2001-825294 3 Apr 2001
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2002-171027 [22]
AB DERWENT ABSTRACT:

NOVELTY - An isolated ovarian tumor polypeptide (I) comprising a sequence (S1) of 55, 67, 73, 787, 453 or 141 amino acids fully defined in the specification, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following: (1) an isolated polynucleotide (II) comprises a sequence selected from: (a) a sequence (S2) selected from 84 sequences having 396 base pairs (bp), and a sequence of 924, 3321, 487, 3999, 1069, 1817, 2382, 2377, 1370, 2060, 3000, 1409, 447, 707, 552, 449, 606, 369, 2008, 2364, 1362, 625, 1619, 1010, 480 or 1897 bp fully defined in the specification; (b) complements of (S2); (c) sequences consisting of at least 20 contiguous residues of (S2); (d) sequences that hybridize to (S2) under moderately stringent conditions; (e) sequences having at least 75% preferably 90% identity to (S2); and (f) degenerate variants of (S2); (2) an isolated polypeptide (III) encoded by (II) comprises a sequence from a sequence (S1); sequences encoded by (II); and sequences having 70% preferably 90% identity to sequence encoded by (II); (3) an expression vector (IV) comprising (II) operably linked to a expression control sequence; (4) a host cell transformed or transfected with (IV); (5) an isolated antibody (Ab), or its antigen binding fragment specific to (III); (6) detecting (M1) an ovarian cancer in a patient, comprising contacting a biological sample from the patient with a binding agent that binds to (III), detecting amount of (III) bound to the binding agent, and comparing the amount to a predetermined cut-off value; (7) a fusion protein (V) comprising (III); (8) an oligonucleotide (OLI) that hybridizes to (S2) under moderately stringent conditions; (9) stimulating and/or expanding (M2) T-cells specific for a tumor protein comprising contacting T-cells with (II), (III) or antigen presenting cells (APC) that express (II); (10) an isolated T-cell population (VI) comprising T-cells prepared by M2; (11) a composition (C1) comprising carriers, immunostimulants, and (I), (II), Ab, (IV), (V) or APC; (12) a diagnostic kit comprising OLI, or Ab and detection reagent comprising a reporter group; and (13) inhibiting (M3) the development of a cancer in a patient comprising incubating CD4+ and/or CD8+ T cells isolated from a patient with (III), (II) or APC, such that T cell proliferate, and administering

to the patient the proliferated T cells.

WIDER DISCLOSURE - Also disclosed are: (1) fragments and/or derivatives of (I); (2) monitoring a progression of a cancer in a patient; (3) polynucleotide compositions comprising antisense oligonucleotides; (4) peptide **nucleic acids** comprising (II); (5) and (5) binding agents specific to (I).

BIOTECHNOLOGY - Preparation: (I) is prepared by standard recombinant techniques.

ACTIVITY - Cytostatic. No biodata is given in the source material.

MECHANISM OF ACTION - Vaccine; Gene therapy.

USE - M1 is useful for detecting a cancer in a patient; M2 is useful for stimulating and/or expanding T-cells specific for a tumor protein; and (M3) is useful for inhibiting the development of a cancer in a patient. C1 is useful for stimulating an immune response in a patient and for treating a cancer in a patient. OLI is useful for determining the presence of a cancer in a patient. The method comprises contacting biological sample from the patient with OLI, detecting amount of (II) that hybridizes to OLI, and comparing the amount to a predetermined cutoff value (claimed). (VI) is further useful for removing tumor cells from a biological sample. (II) is useful for their ability to selectively form **duplex** molecules with complementary stretches of the entire desired gene or gene fragments, and for designing and preparing ribozyme molecules for inhibiting expression of tumor polypeptides in tumor cells. (I), (II), (III) or (V) is useful in recombinant **DNA** molecules to direct expression of a polypeptide in appropriate host cells. Host cells transformed with (II) is useful for preparation of (I).

ADMINISTRATION - C1 comprising (II) is administered by viral based sequence, vaccinia-based infection/transfection system, or is delivered as naked **DNA** or via particle bombardment. C1 is administered through topical, oral, nasal, mucosal, intravenous, intracranial, intraperitoneal, subcutaneous or intramuscular route, or as lipozomes, nanocapsules, **microparticles**, lipid particles or vesicles. Further C1 can also be administered through intraperitoneal, subcutaneous, intradermal, anal, vaginal or topical route. Dosage of (III) is 25 microg-5 mg/kg.

EXAMPLE - Primary ovarian tumor and metastatic ovarian tumor cDNA **libraries** were each constructed in kanamycin resistant pZErO-(RTM) 2 vector from pools of three different ovarian tumor RNA samples. For the primary ovarian tumor **library**, the following RNA samples were used: a moderately differentiated papillary serous carcinoma of a 41 year old, a stage IIC ovarian tumor and a papillary serous adenocarcinoma for a 50 years old Caucasian, and for the metastatic ovarian tumor **library**, the RNA samples used were omentum tissue from: a metastatic poorly differentiated papillary adenocarcinoma with psammoma bodies in a 73 year old, a metastatic poorly differentiated adenocarcinoma in a 74 years old and a metastatic poorly differentiated papillary adenocarcinoma in a 68 year old, where the number of clones in each **library** was estimated by plating serial dilutions of unamplified **libraries**. Insert data were determined from 32 primary ovarian tumor clones and 32 metastatic ovarian tumor clones. Four subtraction **libraries** were constructed in ampicillin resistant pcDNA31 vector. Two of the **libraries** were from primary ovarian tumors and two were from metastatic ovarian tumors. In each case, the number of restriction enzyme cut within inserts was minimized to generate full length subtraction **libraries**. Two hybridizations were performed, and notI-cut pcDNA3.1(+) was the cloning vector for the subtracted **library**. Still further ovarian carcinoma polynucleotide and/or polypeptide sequences identified. Sequences 0574S, 05845 and 0585S represented novel sequences. The remaining sequences exhibited at least some homology with known genomic and/or expression sequence tag (EST) sequences. (131 pages)

L140 ANSWER 3 OF 9 BIOSIS

ACCESSION NUMBER: 2003:117662 BIOSIS
DOCUMENT NUMBER: PREV200300117662
TITLE: Solid phase selection of differentially expressed genes.
AUTHOR(S): Albrecht, Glenn; Brenner, Sydney (1); DuBridge, Robert B.
CORPORATE SOURCE: (1) Cambridge, UK UK
ASSIGNEE: Lynx Therapeutics, Inc.
PATENT INFORMATION: US 6511802 January 28, 2003
SOURCE: Official Gazette of the United States Patent and Trademark Office Patents, (Jan. 28 2003) Vol. 1266, No. 4, pp. No Pagination. http://www.uspto.gov/web/menu/patdata.html. e-file.
ISSN: 0098-1133.
DOCUMENT TYPE: Patent
LANGUAGE: English

AB The invention provides a method and materials for monitoring and isolating differentially expressed genes. In accordance with the method of the invention, differently labeled populations of DNAs from sources to be compared are competitively hybridized with reference **DNA** cloned on solid phase supports, e.g. **microparticles**, to provide a differential expression **library** which, in the preferred embodiment, may be manipulated by fluorescence-activated cell sorting (FACS). Monitoring the relative signal intensity of the different fluorescent labels on the **microparticles** permits quantitative analysis of expression levels relative to the reference **DNA**. The invention also provides a method for identifying and isolating rare genes. Populations of **microparticles** having relative signal intensities of interest can be isolated by FACS and the attached DNAs identified by sequencing, such as with massively parallel signature sequencing (MPSS), or with conventional **DNA** sequencing protocols.

L140 ANSWER 4 OF 9 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER: 2001:428068 BIOSIS
DOCUMENT NUMBER: PREV200100428068
TITLE: Solid phase selection of differentially expressed genes.
AUTHOR(S): Albrecht, Glenn; Brenner, Sydney (1); DuBridge, Robert B.
CORPORATE SOURCE: (1) Cambridge UK
ASSIGNEE: Lynx Therapeutics, Inc.
PATENT INFORMATION: US 6265163 July 24, 2001
SOURCE: Official Gazette of the United States Patent and Trademark Office Patents, (July 24, 2001) Vol. 1248, No. 4, pp. No Pagination. e-file.
ISSN: 0098-1133.
DOCUMENT TYPE: Patent
LANGUAGE: English

AB The invention provides a method and materials for monitoring and isolating differentially expressed genes. In accordance with the method of the invention, differently labeled populations of DNAs from sources to be compared are competitively hybridized with reference **DNA** cloned on solid phase supports, e.g. **microparticles**, to provide a differential expression **library** which, in the preferred embodiment, may be manipulated by fluorescence-activated cell sorting (FACS). Monitoring the relative signal intensity of the different fluoresent labels on the **microparticles** permits quantitative analysis of expression levels relative to the reference **DNA**. Populations of **microparticles** having relative signal intensities of interest can be isolated by FACS and the attached DNAs identified by sequencing, such as with massively parallel signature sequencing (MPSS), or with conventional **DNA** sequencing protocols.

L140 ANSWER 5 OF 9 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER: 2000:279075 BIOSIS
DOCUMENT NUMBER: PREV200000279075
TITLE: Detection of amplified **nucleic acid**

sequences using bifunctional haptenization and dyed **microparticles**.
AUTHOR(S): Gerdes, John C. (1)
CORPORATE SOURCE: (1) Denver, CO USA
ASSIGNEE: Molecular Innovations, Inc., Denver, CO, USA
PATENT INFORMATION: US 5989813 November 23, 1999
SOURCE: Official Gazette of the United States Patent and Trademark Office Patents, (Nov. 23, 1999) Vol. 1228, No. 4, pp. No pagination. e-file.
ISSN: 0098-1133.
DOCUMENT TYPE: Patent
LANGUAGE: English
AB The invention describes an assay for detecting amplified target **nucleic acid** sequences with a visual signal. The sensitivity and specificity of the methodology are based on bifunctional target labeling during the amplification step or subsequent hybridization that generates a bifunctional label. The invention may be used, for example, in the screening of amplicon detection for the purpose of more efficiently screening **libraries**. The invention is also useful to detect **nucleic acid** sequences indicative of a genetic defect or contagious disease when used with the appropriate primers, as well as detect the existence of **nucleic acid** amplification.

L140 ANSWER 6 OF 9 WPIDS (C) 2003 THOMSON DERWENT
ACCESSION NUMBER: 2002-557381 [59] WPIDS
DOC. NO. CPI: C2002-158089
TITLE: New biocompatible and biodegradable polymer compounds for preparing drug delivery device, pharmaceutical composition, polynucleotide/polymer complex.
DERWENT CLASS: A23 A96 B04 D16
INVENTOR(S): AMIJI, M M; ANDERSON, D G; LANGER, R S; LYNN, D M; PUTNAM, D
PATENT ASSIGNEE(S): (AMIJ-I) AMIJI M M; (ANDE-I) ANDERSON D G; (LANG-I) LANGER R S; (LYNN-I) LYNN D M; (PUTN-I) PUTNAM D; (MASI) MASSACHUSETTS INST TECHNOLOGY
COUNTRY COUNT: 22
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2002031025	A2	20020418	(200259)*	EN	133
RW: AT BE CH CY DE DK ES FI FR GB GR IE IT LU MC NL PT SE TR					
W: CA JP					
US 2002131951	A1	20020919	(200264)		

APPLICATION DETAILS:

PATENT NO	KIND		APPLICATION	DATE
WO 2002031025	A2		WO 2001-US31270	20011005
US 2002131951	A1	Provisional	US 2000-239330P	20001010
		Provisional	US 2001-305337P	20010713
			US 2001-969431	20011002

PRIORITY APPLN. INFO: US 2001-969431 20011002; US 2000-239330P 20001010; US 2001-305337P 20010713
AB WO 200231025 A UPAB: 20021031
NOVELTY - Polymer compounds (I)-(VIII) are new.
DETAILED DESCRIPTION - Polymer compounds of formulae (I)-(IV) and (V)-(VIII) and their derivatives and salts are new.
A1, B1 = linker of 1-30C carbon chain or 1-30C **heteroatom**

containing carbon chain, (substituted by at least one of alkyl, alkenyl, alkynyl, amino, alkylamino, dialkylamino, trialkylamino, aryl, ureido, (aromatic) **heterocyclic**, (aromatic) cyclic, halogen, hydroxyl, alkoxy, cyano, amide, carbamoyl, carboxylic acid, ester, carbonyl, carbonyldioxyl, alkylthioether or thiol); or

L1 = absent, single bond or a linker group additionally bonded to A1 to form a cyclic structure;

R1-R10 = H, alkyl, alkenyl, alkynyl, aryl, halogen, hydroxyl, alkoxy, carbamoyl, carboxyl ester, carbonyldioxyl, amide, thiohydroxyl, alkylthioether, amino, alkylamino, dialkylamino, trialkylamino, cyano, ureido, substituted alkanoyl, cyclic, aromatic cyclic, **heterocyclic**, aromatic **heterocyclic** (all optionally substituted by alkyl, alkenyl, alkynyl, amino, alkylamino, dialkylamino, trialkylamino, aryl, ureido, (aromatic) **heterocyclic**, (aromatic) cyclic, halogen, hydroxyl, alkoxy, cyano, amide, carbamoyl, carboxylic acid, ester, carbonyl, carbonyldioxyl, alkylthioether or thiol);

M1 = 1,3-imidazolidinyl or a group of formula (a)-(c);

M' = (CH2)2CH(CH3) or (CH2)4;

S1 = 4-hydroxybutyl, 2-(2-hydroxyethoxy)ethyl, 3-(dimethylamino)propyl or 3-morpholinopropyl;

X- = chloride, fluoride, bromide, iodide, sulfate, nitrate, fumarate, acetate, carbonate, stearate, laurate or oleate;

n = 5-10000;

m = 0-50; and

a = 3-10000.

INDEPENDENT CLAIMS are included for the following:

(1) composition comprising polynucleotide and polymer compound;

(2) synthesizing poly(beta -amino ester) (PBAE) by reacting bis-(secondary amine) and bis(acrylate ester under suitable conditions;

(3) encapsulating an agent in poly(beta -amino ester) matrix to form **microparticles** by contacting agent and PBAE under suitable condition to form **microparticles**; and

(4) screening a **library** of polymers which involves providing several polymers and screening polymers for desired property useful in gene therapy.

USE - For preparing drug delivery device, pharmaceutical composition, nanometer scale complexes with **nucleic acid**, polynucleotide/polymer complexes, encapsulation of therapeutic diagnostic and/or prophylactic agent e.g. polynucleotides to form **microparticles**.

ADVANTAGE - Polymer is biodegradable and biocompatible.

DESCRIPTION OF DRAWING(S) - The figure shows degradation, expressed as % degradation over time based on GPC data, of polymers (1-3) at 37 deg. C and pH 5.1, 7.4.

Dwg.1/10

L140 ANSWER 7 OF 9 WPIDS (C) 2003 THOMSON DERWENT

ACCESSION NUMBER:	2002-646964 [70] WPIDS
DOC. NO. NON-CPI:	N2002-511705
DOC. NO. CPI:	C2002-182586
TITLE:	Selecting a biochemical substance by the surface presentation method which utilizes a material A with specific affinity for the material B, where material A is immobilized to a particle.
DERWENT CLASS:	A89 B04 D16 S03
PATENT ASSIGNEE(S):	(TTOC) TOTO LTD
COUNTRY COUNT:	1
PATENT INFORMATION:	

PATENT NO	KIND	DATE	WEEK	LA	PG
JP 2002071692	A	20020312	(200270)*		7

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
JP 2002071692	A	JP 2000-226374	20000727

PRIORITY APPLN. INFO: JP 2000-174644 20000612

AB JP2002071692 A UPAB: 20021031

NOVELTY - Selecting (M1) a biochemical substance, which is material B, by the surface presentation method which utilizes a material A with a specific affinity for the material B, where material A is immobilized to a particle.

DETAILED DESCRIPTION - An INDEPENDENT CLAIM is included for a selection kit (I) used in the case of a target substance is from the **library** containing a target substance, with the **microparticle** and at least a reaction container.

USE - (M1) is useful for selecting biochemical substance, by the surface presentation method (claimed).

Dwg.1/4

L140 ANSWER 8 OF 9 WPIDS (C) 2003 THOMSON DERWENT

ACCESSION NUMBER: 2000-365646 [31] WPIDS
DOC. NO. CPI: C2000-110508
TITLE: Synthesizing oligonucleotide tags for use in specific hybridization processes.
DERWENT CLASS: B04 D16
INVENTOR(S): DUBRIDGE, R B; KIRCHNER, J J; WILLIAMS, S R
PATENT ASSIGNEE(S): (LYNX-N) LYNX THERAPEUTICS INC
COUNTRY COUNT: 90
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2000026411	A1	20000511	(200031)*	EN	43
AU 2000016031	A	20000522	(200040)		
EP 1127161	A1	20010829	(200150)	EN	
JP 2002528137	W	20020903	(200273)		73

WO 2000026411 A1 designated states:
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW NL OA PT SD SE SL SZ TZ UG ZW
W: AE AL AM AT AU AZ BA BB BG BR BY CA CH CN CR CU CZ DE DK DM EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT TZ UA UG US UZ VN YU ZA ZW

EP 1127161 A1 designated states:
R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT RO SE SI

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2000026411	A1	WO 1999-US25680	19991101
AU 2000016031	A	AU 2000-16031	19991101
EP 1127161	A1	EP 1999-958733	19991101
		WO 1999-US25680	19991101
JP 2002528137	W	WO 1999-US25680	19991101
		JP 2000-579783	19991101

FILING DETAILS:

PATENT NO	KIND		PATENT NO
AU 2000016031	A	Based on	WO 200026411

EP 1127161 A1 Based on WO 200026411
JP 2002528137 W Based on WO 200026411

PRIORITY APPLN. INFO: US 1998-106662P 19981102
AB WO 200026411 A UPAB: 20000630

NOVELTY - A method (I) for the synthesis of oligonucleotide tag collections, comprising producing a repertoire of oligonucleotide tag complements on a solid support, cleaving the tag complements from the solid support and inserting the cleaved components into a cloning vector, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are included for the following:

(1) a method (I) of synthesizing a repertoire of oligonucleotide tags, comprising:

(a) cleaving a number of different sequence oligonucleotide populations one solid phase supports so that each population is located at a spatially discrete region relative to other populations and each population comprise a tag-complement sequence that is the same for every oligonucleotide in a given population;

(b) cleaving a fraction of the oligonucleotides from each population on the supports to release a mixture of different-sequence tag-complement oligonucleotides; and

(c) annealing a primer to each tag-complement oligonucleotide and extending each annealed primer to form a **duplex** comprising a tag-complement strand and a complementary tag strand, so that the tag-complement strands or the **duplexes** formed comprise an oligonucleotide tag repertoire;

(2) a repertoire of tag oligonucleotides (II) prepared by (I);

(3) a system (III) for sorting polynucleotides, comprising:

(a) one or more solid phase supports to which a number of oligonucleotide populations are attached so that each population is located at a spatially discrete region relative to other populations and each population comprise a tag-complement sequence that is the same for every oligonucleotide in a given population; and

(b) a tag composition comprising (II) prepared from the solid supports; and

(4) a solid phase bead (or beads) (IV) comprising a population of attached oligonucleotides, which comprises 2 classes of oligonucleotide one of which comprises a cleavable linking group that permits the selective cleavage from the support without cleaving the second class of oligonucleotides.

USE - The oligonucleotide tags produced may be used in specific hybridization procedures for identifying, sorting and tracking molecules, especially polynucleotides, in a range of research, industrial and medical applications, e.g. the identification of disease related polynucleotides in diagnostic assay, screening for clones of novel target oligonucleotides, identification of specific polynucleotides in blots of mixtures of polynucleotides, amplification of target polynucleotides and therapeutic blocking of inappropriately expressed genes (for example, see Sambrook et al., Molecular Cloning: A Laboratory Manual (1989) and Keller et al., **DNA** Probes (1993)).

ADVANTAGE - The cloned tag complements can be conventionally be conjugated to selected polynucleotides to produce tagged polynucleotides with unique tag sequences which can be captures and sorted by hybridization to corresponding tag complements.

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